Effects of exercise on hunger, food intake and energy expenditure

ROCHA, Joel Borges Pinto Ferreira da

REFERENCE

Effects of exercise on hunger, food intake and energy expenditure

Joel Borges Pinto Ferreira da Rocha

A doctoral thesis submitted in partial fulfilment of the requirements of Sheffield Hallam University for the degree of Doctor of Philosophy

June 2013

Abstract

Research in this thesis has examined the acute and chronic effects of exercise on hunger, energy intake and expenditure. Cross-sectional studies examined the effect of 60 min of moderate-intensity cycling on immediate and subsequent three day energy intake and expenditure in active and inactive men (study one) and women not using hormonal contraceptives (study two) and taking oral contraceptives (study three). Study four examined the effects of 12 weeks of moderate-intensity aerobic exercise on 7-day free-living energy intake and expenditure. A total of 47 men (mean ± SD; age 23.8 ± 4.2 y; body mass index 24.2 ± 3.0 kg-m'2) and 52 women (22.7 ± 3.4 y; 22.1 ± 2.1 kg-m'2) were recruited into four studies.

In study one, 60 min of moderate-intensity (50% of maximum oxygen uptake) cycling did not have an effect on hunger or *ad libitum* lunch energy intake *(p* > 0.05) but induced an acute (within the experimental day, *p* = 0.024, *d* = 0.56) and delayed (third day after the experimental day,/> = 0.024, <i= 0.80) increase in free-living energy intake in active and inactive participants, respectively with no compensatory changes in free-living energy expenditure *(p* > 0.05). Similarly, studies two and three demonstrated that an acute bout of moderate-intensity aerobic exercise does not increase hunger or *ad libitum* lunch energy intake in active and inactive women *{p* > 0.05). In study two there were no exercise-induced compensatory responses in free-living energy intake *{p* > 0.05) whereas in study three, the inactive group decreased their daily energy intake on the first day after the exercise experimental day compared with control *(p* = 0.002, *d* = -0.89). No compensatory changes in daily physical activity energy expenditure were observed in these studies *(p* > 0.05). In study four 12 weeks of moderate-intensity aerobic exercise did not induce changes to weekly free-living energy intake and expenditure *(p* > 0.05) despite the high inter-individual variability in changes in body composition. Additionally, inactive participants are not able to independently maintain their physical activity behaviour after the end of a supervised exercise intervention.

Overall, this research shows that an acute bout of moderate-intensity aerobic exercise did not affect hunger irrespective of sex or habitual physical activity, however the use of oral contraceptives may have heightened appetite in women. Active men were able to compensate for the acute exercise-induced energy deficit by increasing their energy intake quicker (within the experimental day) than inactive men (third day after the experimental day). In women, no clear relationship was apparent. Moreover, an acute bout of exercise did not elicit compensatory changes in physical activity in men and women. These findings enhance the knowledge of how an acute bout of exercise affects immediate and subsequent energy intake and expenditure in active and inactive men and women but more work is needed to confirm and explore the potential causal mechanisms.

Acknowledgements

This PhD thesis is the result of a challenging journey, that would not have been possible without the help and support of many people.

My first debt of gratitude must go to all my supervisors, Dr David Broom, Jenny Paxman and Dr Caroline Dalton for giving me the opportunity to undertake this PhD. Their guidance, encouragement and support helped me in all the time of research and writing of this thesis.

I would also like to thank my director of studies, Prof. Edward Winter, that contributed to my continuous development particularly on the skill of writing and knowledge of statistics.

I am very grateful to all the technicians at Collegiate Hall and the 12th floor of Owen Building. Completing my testing at two sites was often challenging, and it would certainly not have been possible without their help, support and friendly environment.

The friendships I made during this journey were also very important to me. In particular, sharing offices with James and Steve, and later Joe has, without any doubt, made this a more enjoyable experience. I will definitely miss the constant banter and exchange of ideas. So for these dear friends I want to express my deepest gratitude.

I am also sincerely grateful to all the volunteers who willingly gave up their time to participate in my studies. I can honestly say that without all your help this would not have been possible.

Despite not being involved in my PhD, Dr. David Stensel was the person who encouraged and inspired me to pursue it. Therefore, I want to sincerely thank him for being such a positive influence during my first year in the United Kingdom.

Finally, I am hugely indebted to my family and therefore I dedicate this thesis for all of them. I wish to thank my mother, my auntie Isabel and my grandmother Maria for all their love and inspiration. To all my sisters, the people who I most admire and love in this world, I can only say a heartfelt "thank you". They are a constant warm presence in my heart that keeps me going even through the hardest times. To my father, whose life was too short to see me grow up and become a man, I can only hope that this work makes him proud. Last but not least, I want to thank Joana for her constant love, friendship and kindness.

Statement of original authorship

I declare that the work contained in this thesis is original, and is my work except where otherwise stated. The material of other authors has been cited with their names and source of publication.

Publications and presentations arising from this thesis

Submitted article

Effects of an acute bout of aerobic exercise on immediate and subsequent three-day food intake and energy expenditure in active and inactive men. *Appetite*

Presentations and conference proceedings

Oral presentation "Effects of an acute bout of moderate-intensity aerobic exercise on subsequent 3-day food intake and energy expenditure in active and inactive males" at the Nutrition Society Postgraduate Conference in Newcastle, September 2012.

Poster presentation "Effects of an acute bout of exercise on hunger, energy intake and expenditure in active men" - at the Health and Wellbeing Faculty Research Day (Sheffield Hallam University), June 2011.

Oral presentation of PhD project "Effects of exercise on hunger, food intake and energy expenditure" - at the Centre of Sport and Exercise Science Meeting (Sheffield Hallam University), December 2010.

Table of contents

List of figures

Chapter 6

List of tables

Chapter 6:

Chapter 7:

List of abbreviations

95% Cl	95% confidence interval
ACSM	American College of Sports Medicine
AgRP	agouti-related peptide
AUC	Area under the curve
BIA	Bioelectrical impedance analysis
BMI	Body mass index
BMR	Basal metabolic rate
CART	Cocaine and amphetamine related transcript
CCK	Cholecystokinin
CHO	Carbohydrates
EE	Energy expenditure
El	Energy intake
FCI	Food Craving Inventory
FFM	Fat free mass
FM	Fat mass
GLP-1	Glucagon-like peptide 1
GLTEQ	Godin Leisure-Time Exercise Questionnaire
GLUT-4	Glucose transporter type 4
HDL	High density lipoproteins
HR	Heart rate
HRR	Heart rate reserve
ICC	Intraclass correlation coefficient
LDL	Low density lipoproteins
NEAT	Non-exercise activity thermogenesis
NPY	Neuropeptide Y
OC	Oral contraceptives
PAEE	Physical activity energy expenditure
PAL	Physical activity level
POMC	Proopiomelanocortin
PYY	Peptide tyrosin-tyrosin
R	Coefficient of reliability

REI	Relative energy intake
RER	Respiratory exchange ratio
RPE	Rating of perceived exertion
SD	Standard deviation
SEM	Standard error of the mean
SIPM	Sussex Ingestion Pattern Monitors
SMM	Skeletal muscle mass
SPAF	Shortened Premenstrual Assessment Form
TC	Total cholesterol
TE	Typical error
TEE	Total energy expenditure
TEF	Thermic effect of food
TEM	Technical error of measurement
TFEQ-R18	Revised version of the Three-Factor Eating
VAS	Visual analogue scale
$V02max$	Maximum oxygen consumption
VFA	Visceral fat area
VTA	Ventral tegmental area
a-MSH	Alpha-melanocyte stimulating hormone

Chapter 1: Introduction

1.1 Physical activity, exercise and health

1.1.1 History of physical activity, exercise and health

Physical activity has always been a topic of much interest and its relation with health promotion and disease prevention can be traced back to more than 2,000 years ago, when physicians such as Susruta (c. 600 B.C.), Hippocrates (c. 460 - 377 B.C.) and Galen (A.D. 129 - *c.* 216) were already prescribing physical activity and exercise for the prevention and treatment of diseases (Haslam, 2007; Tipton, 2008). This ancient knowledge that exercise improves health, may have been based on observational skills, experiments with condemned criminals, or pure inspiration (Porter, 1999), nevertheless, it still stands today as emerging scientific evidence suggests that regular physical activity reduces the risk of all-cause mortality, cardiovascular disease, stroke, hypertension, colon cancer, breast cancer, type 2 diabetes, osteoporosis, depression and dementia (Kesaniemi *et al.*, 2010).

1.1.2 Definition of physical activity and exercise

According to Caspersen *et al.* (1985) physical activity is defined "as any bodily movement produced by skeletal muscles that results in energy expenditure" and can be divided into subcategories according to the portion of daily life during which the activity occurs. These are occupational physical activity (activity carried out in the workplace), leisure-time physical activity (activities undertaken during the individual free time, i.e. outside occupational work), and sleeping physical activity (physical activity done during sleep). As the latter is negligible for most purposes only the first

two are often considered. The term exercise is a subcategory of leisure-time physical activity and is defined as "planned, structured and repetitive bodily movements that are performed to improve or maintain one or more components of physical fitness" (Caspersen *et al.,* 1985).

Despite the previously presented definitions of physical activity and exercise, these terms are often used interchangeably. According to some authors (Winter & Fowler, 2009), there is little if any practical distinction between the two terms with the difference being determined by an interpretation of the motivation or intent of the participant which could give rise to one person's exercise being another person's physical activity. In addition, they demonstrated that Caspersen *et al.* (1985) definitions of physical activity and exercise were flawed as they do not recognise that they do not always require movement (e.g. isometric muscle activity) to induce energy expenditure. According to Winter and Fowler (2009), both terms should be defined as "a potential disruption to homeostasis by muscle activity that is either exclusively, or in combination, concentric, eccentric or isometric".

Precise universal definitions of terms with such subtle differences are difficult to achieve, however, a distinction between the two terms allows a better understanding of the type of activity that is being referred to. Thus, in this thesis the term physical activity will include any daily living activity that involves either exclusively, or in combination, concentric, eccentric or isometric muscle activity while exercise will be used as a more specific term that implies planned, structured, repetitive and purposeful physical activity.

1.2 Obesity

1.2.1 Definition and prevalence of obesity

Obesity is a multi-factorial condition, characterised by the excess accumulation of body fat, associated with various health problems that diminish quality of life and limit lifespan. It presents an economic burden that includes costs of chronic disease management and loss of work productivity (Eckel, 2008a). The prevalence of obesity has greatly increased over the past 30 years and is considered one of the most profound public health challenges of our time affecting not only the developed world but also developing nations (Caballero, 2007). Globally, approximately 1.46 billion adults (>20 years) are classified as overweight (BMI >25 kg/m2) and at least 400 million as obese (BMI >30 kg/m) (Finucane *et al*., 2011). In England, the prevalence of obesity in adults has almost doubled in the last 20 years and the proportion of obese adults has increased from 1993 to 2011 from 13% to 24% in men and 16% to 26% in women (Sutton, 2012).

1.2.2 Causes of obesity

Albeit that genetic factors are important in determining an individual's susceptibility to an increase in body mass, it is generally agreed that the rapid worldwide increase in obesity is not attributable to genetic factors alone because it is unlikely that the human gene pool has recently changed (WHO, 2000). Environmental and behavioural changes are therefore likely to be the main factors behind this rapid increase of obesity in recent years (Hill & Peters, 1998). Despite developing from the complex interactions of factors such as the quality and quantity of dietary intake, environmental and genetic inputs and physiological and psychological status, obesity results from a prolonged state of

positive energy balance (Swinbum *et al.,* 2011) that can be attributable to an increase in energy intake, a decrease in energy expenditure or a combination of both.

1.2.3 Obesity management strategies

Current obesity management strategies include individual approaches such as exercise interventions, dietary modification, behavioural therapy, pharmacological treatments and surgery, or a combination of these for greater efficacy. While the non-surgical strategies are often the first and preferred approach, surgery is the recommended and most effective treatment for morbidly obese patients (BMI > 40 kg/m2) or for severely obese individuals (> 35 kg/m) if in the presence of marked comorbidities in which surgically-induced body mass reduction is expected to improve the disorder (Fried *et al.,* 2007). Bariatric surgery can be used to achieve a sustained reduction of body mass and the resolution or improvement of obesity-related comorbidities (diabetes, hyperlipidaemia, hypertension and obstructive sleep apnoea), however, it is important to acknowledge that it can be associated with certain perioperative risks and late complications (Pories, 2008).

Pharmacological treatments (e.g. Orlistat: an agent that inhibits activity of pancreatic and gastric lipases and is able to induce reductions in body mass by decreasing the gastrointestinal absorption of ingested fat by approximately 30%) (Drent *et al.,* 1995) (are another strategy used to facilitate a reduction in body mass and similarly with bariatric surgery there are criteria necessary to prescribe these treatments to patients. The prescription of medications is normally considered if lifestyle modifications such as diet and physical activity have proved ineffectual and requires the individual to have a BMI greater than 30 kg/m^2 or at least 27 kg/m^2 when obesity-related comorbidities are

present (Glandt & Raz, 2011). Despite being able to induce a reduction of body mass compared with placebo after 1 year, pharmacological treatments are regarded as a frequent adjunct to other strategies as the decrease in body mass remains modest (e.g. 95% Cl, -2.5 to -3.2 kg for Orlistat compared with placebo at > 12 months) and is not sustained when therapy is discontinued with most patients regaining some or all the body mass lost originally (Kang & Park, 2012). Moreover, patient compliance, cost of medication and possible side effects are other factors that can make this strategy difficult to implement (Ioannides-Demos *et al.,* 2011).

Behavioural therapy is an approach used to help individuals develop a set of skills to achieve a healthier body mass and includes goal setting, self-monitoring, stimulus control (identification and modification of the patient's environment to enhance behaviours that will support the management of body mass), cognitive restructuring (increasing the awareness of the perceptions of themselves and their body mass), stress management and social support (Poston & Foreyt, 2000). This strategy that often involves nutrition and physical activity counselling, is delivered either individually or in small groups and has been reported to produce a body mass reduction of 8 to 10% during the first six months of treatment (Foster *et al.,* 2005). Despite these encouraging values it is important to consider that most studies have been undertaken in academic medical centres and the success of these strategies in other treatment settings is less clear (Eckel, 2008b). Additionally, access and adherence to this approach may be often limited by the cost and availability of these programs, relationship with the patients, transportation concerns and time constraints (Wadden *et al.,* 2007).

Dietary modification is an obesity management strategy that directly targets energy intake. Treatment options include stabilisation of eating (correcting erratic eating patterns), improvement of the diets quality (e.g. decrease saturated fat), implementation of reduced-energy diets, low-glycaemic index diets and meal replacements (vitamin and mineral fortified products that replace one or two meals per day) (Grace, 2011). From all these treatments, reduced-calorie diets remains one of the most applied treatments which aim to induce very low (less than 800 kcal / 3.35 MJ daily), low (800 to 1500 kcal / 3.35 to 6.28 MJ daily), or moderate (about 500-600 kcal / 2 09-2-51 MJ less than typical daily intake) daily energy intakes. From these, the latter is often the recommended starting treatment as the energy deficit of 500-600 kcal (2-09-2-51 MJ) has been comprehensively implemented in long-term trials and identified in Cochrane analyses as one of the best options (Haslam & James, 2005). Additionally, very-low-energy diets require medical monitoring because they can further increase the drive to eat putting heavier patients under greater physiological stress (Eckel, 2008b). The efficacy of the different dietary treatments remains difficult to assess because of heterogeneous samples, types of treatment and follow-up time, however, in a systematic review of randomized controlled trials, Heymsfield *et al.* (2003) reported that, at one year, meal replacements induced a 7-8% body mass reduction while conventional reduced-calorie diets induced only 3-7%. In a review of 51 diet alone interventions, the decrease of body mass at 6, 12, 24 and 36 months averaged 5%, 4.6%, 4.4% and 3.0% respectively showing that in the long-term there is often a regain of at least one-third of the body mass initially lost over 1 year (Franz *et al*., 2007). Moreover, recent studies (Sacks *et al.,* 2009; De Souza *et al.,* 2012) comparing different diets that promote the reduction of body mass reported that the reduction in total energy intake, rather than modification of the macronutrient content of the diet, was the most important

determinant of fat loss, therefore the best diet remains the one that the individual will best and longest adhere to. Despite the beneficial health outcomes that have been reported in the short-term (Hensrud, 2012) there are a number of safety concerns about the long term use of dietary treatments alone such as the loss of fat free mass, psychological distress (e.g., preoccupation with shape and "weight"), possible health hazards of body mass cycling, and increased risk of developing eating disorders (Brownell & Rodin, 1994).

Despite exercise interventions alone, without decreased caloric intake, being associated with only a modest body mass reduction (Wing, 1999), exercise is still one of the best recognised obesity prevention and management strategies. The importance of exercise in interventions that aim to reduce body mass has been traditionally associated to its ability to bum calories and the potential to generate a state of negative energy balance (Jakicic, 2002). However, there is a large body of evidence supporting that regular exercise contributes to the prevention of some diseases and promotes metabolic adaptations that improve physical and mental health (Bertheussen *et al*., 2011; Chaput *et al.*, 2010; Tremblay & Therrien, 2006). As with dietary interventions, the efficacy of exercise interventions in generating a decrease or preventing an increase in body mass within a period of time is difficult to assess due to factors such as the variable adherence to the exercise prescribed, its characteristics (type, duration, frequency, and intensity), ability to modify post-exercise energy metabolism, and the nutritional context surrounding its practice (Tremblay & Therrien, 2006). Nevertheless, a review of 16 randomised controlled trials using exercise alone interventions (duration ranged between 4 and 16 months and prescription -60-180 min per week) reported a body mass reduction ranging from 0.1-5.2 kg, with most studies showing a reduction of only

1-3 kg (Catenacci & Wyatt, 2007). A Cochrane review of 41 randomised controlled trials (duration ranged between 3 and 12 months and prescription -45-360 min per week) also reported that exercise alone interventions induced a body mass reduction ranging between 0.5 to 4.0 kg (Shaw *et al.,* 2006). In this review, exercise intensity was a strong determinant of body mass reduction with high-intensity exercisers losing about 1.5 kg more than low-intensity exercisers. Despite the limited ability that exercise has to induce a reduction of body mass in the short-term, when carefully controlled and maintained for more than 12 months, exercise has a stronger effect on body mass (Donnelly *et al.,* 2003). Moreover, it favourably alters body composition (Elder & Roberts, 2007), health-related outcomes (Jakicic & Otto, 2006) and is associated with long-term success in the maintenance of reduced body mass (Wing & Phelan, 2005). Regarding the latter association, it is still not clear why this is the case (Hill & Wyatt, 2005) but effects on energy intake (Martins *et al.,* 2008) or other components of energy expenditure, including resting metabolic rate (RMR) and spontaneous physical activity (Catenacci & Wyatt, 2007) are a possibility.

1.3 Summary

Exercise, when properly managed, is probably the best individual obesity management treatment and an essential element to be incorporated into every combined strategy intended to prevent an increase or induce a decrease in body mass (Chaput *et al.,* 2010). Therefore, a better understanding of how exercise impacts energy homeostasis and the regulation of body mass may help the development of better strategies to combat the recent worldwide increase in the prevalence of obesity and its associated morbidity and mortality. For this reason, this thesis presents a body of research that will enhance the

knowledge of the immediate and delayed effects that an acute bout of aerobic exercise has on food intake and energy expenditure in active and inactive men and women and the effects of a 12-week exercise intervention on a week's energy intake and energy expenditure in inactive men.

1.3.1 Thesis overview

This thesis comprises four original studies organised by chapters. Study one (Chapter 4), examined effects of an acute bout of moderate intensity aerobic exercise on immediate and subsequent three day food intake and energy expenditure in active and inactive men. Study two (Chapter 5) examined effects of an acute bout of moderate intensity aerobic exercise on immediate and subsequent three day food intake and energy expenditure in active and inactive women not using any hormonal contraceptives. Study three (Chapter 5) examined effects of an acute bout of moderate intensity aerobic exercise on immediate and subsequent three day food intake and energy expenditure in active and inactive women taking oral contraceptives. Finally, study four (Chapter 6) examined effects of a 12 week exercise intervention on 7 day energy intake and energy expenditure in inactive men.

1.3.2 Research questions

Chapter 4 - Study 1: Does an acute bout of aerobic exercise have an effect on immediate and subsequent three-day food intake and energy expenditure in active and inactive men compared with a resting condition?

Chapter 5 - Study 2: Does an acute bout of aerobic exercise have an effect on immediate and subsequent three-day food intake and energy expenditure in active and inactive women not taking any hormonal contraceptives compared with a resting condition?

Chapter 5 - Study 3: Does an acute bout of aerobic exercise have an effect on immediate and subsequent three-day food intake and energy expenditure in active and inactive women on oral contraceptives compared with a resting condition?

Chapter 6 - Study 4: Does a 12-week aerobic exercise intervention have an effect on 7-day energy intake and energy expenditure in inactive men?

Chapter 2: Literature review

2.1 Chapter overview

This chapter will address the literature on the effects of exercise on hunger, energy intake and physical activity energy expenditure. The chapter starts by describing theories about the regulation of body mass and examining the concept of energy balance and its components. Thereafter, there is a review of early studies ontherelationship between energy intake and expenditure followed by the definitions of, and factors influencing, appetite and physical activity. Finally, there will be a discussion of the relationship between physical activity and appetite, and the literature on the effects of acute and chronic exercise on hunger, energy intake and physical activity energy expenditure. The aims of the studies presented within this thesis will also be listed in the end of this chapter.

2.2 Regulation of body mass

The maintenance of an adequate body mass and composition is considered a major determinant of the survival of mammals (Jequier & Tappy, 1999). In humans, this stability can be seen in most healthy individuals as they are able to maintain a relatively constant body mass through their adult life despite the daily fluctuations in energy intake and expenditure (Goran, 2000). This observation supports the presence of a biological homeostatic mechanism for the regulation of body mass and energy stores.

Based on this homeostatic regulation framework, several theories have been proposed over the years such as the "set-point" (Mrosovsky & Powley, 1977), "settling-point"

(Wirtshafter & Davis, 1977) and the "settling zone" (Levitsky, 2002) models. The "set-point" theory evolved from the hypotheses that circulating factors of specific nutrients that are generated in proportion to body fat stores and/or nutritional status, i.e. fatty acids and glycerol (lipostatic hypothesis) (Kennedy, 1953), glucose (glucostatic hypothesis) (Mayer, 1955) or essential amino acids (aminostatic hypothesis) (Mellinkoff *et al.,* 1956), acted as signals to the brain and/or peripheral organs (e.g. liver) that would elicit changes in feeding behaviour to restore homeostasis. According to the "set-point" theory there is a reference value (set-point) for body fat and by extension body mass which is maintained through an active feedback mechanism linking the body energy stores and energy intake and expenditure (Mrosovsky & Powley, 1977). Thus, this feedback system senses differences between the information of the signal and the reference value and translates it into effects on energy intake and expenditure to maintain homeostasis (Harris, 1990).

Despite being able to incorporate the concept of a biological homeostatic regulation and explain why people often regain the lost body mass once dieting stops (Anderson *et al.,* 2001), the "set-point" theory does not explain the increasing prevalence of obesity and/or allow for environmental factors that influence food intake and physical activity. An alternative "settling point" model proposed by Wirtshafter and Davis (1977), suggested that instead of an active feedback system defending a "set point", body mass would change to a new "settling point" through a passive feedback system. According to this model body mass fluctuates until it "resettles" at a new point defined by the altered energy component (i.e. energy intake or expenditure) (Davis & Wirtshafter 1978). An advantage of this model over the "set-point" theory is its ability to incorporate the environmental influences on obesity such as the availability of energy-dense foods and

the reduction of physical activity (Hill & Peters, 1998). However, the passive feedback system works only on the assumption that one energy component is unregulated and the other is regulated (e.g. if energy stores are affected by an unregulated change in energy intake, energy expenditure will regulate until a new balance is achieved). This assumption conflicts with recent evidence supporting that the brain controls food intake and energy expenditure in response to central and peripheral signals that provide information regarding the body's nutritional status (Murphy & Bloom, 2006; Woods & D'Alessio, 2008; Zhang *et al.*, 1994).

Through acknowledging the limitations of previous theories, Levitsky (2002) proposed the alternative "settling zone" model, which suggests that the "zone" within which body mass is maintained stable may be determined biologically but the behaviours controlling energy intake and expenditure are primarily influenced by environmental and cognitive factors. Thus, there are upper and lower limits that define the points at which the biological regulation is determinant, and between which there is only a small or no biological regulation of body mass (Levitsky, 2002). This model has the advantages of incorporating both the environmental and biological factors affecting the body energy stores and providing a context of understanding the asymmetry of the regulation of body mass (i.e. a reduction in body mass is strongly defended but an increase in body mass is not). The variation of the two limits defining the "settling zone" would explain why in the presence of certain environmental stimuli some people are more resistant to losing body mass (lower limit) and/or prone to gaining body mass (upper limit). Despite all these advantages over previous models it is however important to consider that it is currently impossible to know how realistic these model assumptions are (Speakman, 2007).

2.3 Energy balance

The regulation of body mass complies with the first law of thermodynamics, i.e. that energy can be transformed but not created or destroyed (Jequier & Tappy, 1999). Therefore, to understand how body mass is regulated it is important to acknowledge the concept of energy balance. Assuming that an individual has no problems in absorbing nutrients, energy balance is often calculated as follows:

Energy balance = Energy intake (El) - Energy expenditure (EE)

In this equation energy balance represents the change in body energy stores which is calculated by the difference between energy intake (food and drink consumption) and energy expenditure (physical activity and metabolic processes) (Spiegelman & Flier, 2001). If energy intake equals energy expenditure a state of energy balance is achieved, hence no changes in body mass occur. However, if there is a sustained imbalance between energy intake and expenditure, body mass will change. A prolonged state of positive energy balance (energy intake exceeds expenditure) will lead to an increase in body mass while a prolonged state of negative energy balance (energy expenditure exceeds intake) will lead to a decrease in body mass.

Although it seems intuitively valid, because when there are no changes in body stores energy intake equates to energy expenditure (Swinbum & Ravussin, 1993), this energy balance equation has been shown to be too static to reflect the energy dynamics of living organisms (Alpert, 1990). Therefore, a dynamic model of energy balance should be used as follows:

$$\text{Rate of change in body energy stores} = \text{Rate or energy intake} - \text{Rate of energy expenditure}$$

The inclusion of "rate" in this equation introduces the time dependency of the components of body mass which takes into consideration effects such as the ones that energy stores have on energy expenditure (e.g. fat-free mass influence on resting metabolic rate) (Alpert, 1990; Swinbum & Ravussin, 1993). This allows this model to reflect the dynamics of the energy fluctuation and respective impact on body mass regulation which makes the classic static model inadequate when considering that it focuses on the acute changes in its components, instead of their chronic states. This dynamic model also explains why small initial increases in energy intake sustained over a number of years do not lead to large increases of body mass as often claimed because individuals will re-establish energy balance with a higher energy intake, expenditure and body stores (Hall *et al.*, 2011).

While these equations provide insight into the understanding of energy balance (Galgani & Ravussin, 2008), its concept cannot be reduced to a simple matter of addition and subtraction because their energy components are constantly changing, interdependent and influenced by many factors (Donnelly & Smith, 2005; Swinbum & Ravussin, 1993). Thus, energy balance depends not only on the time frame over which it is being considered, but also on the control and interactions of its energy components (i.e. body energy stores, energy intake and expenditure).

2.3.1 Body energy stores

The body energy stores comprise proteins, carbohydrates and fat. Body proteins take several forms and are a major constituent of the structural tissues and enzymes in the body, thus they are not used as a primary source of energy unless there is a shortage of carbohydrates or fats (Heymsfield *et al.,* 1982; Speth & Spielman, 1983). Protein stores are associated with water and are believed to be tightly controlled on a day to day basis (Abbott *et al.,* 1988). Carbohydrates are stored primarily as intracellular glycogen in the skeletal muscle and liver and their storage capacity is limited in humans to approximately 15 g/kg of lean body mass (Acheson *et al.,* 1988). Similarly to proteins, glycogen is also stored in association with water which reduces its energy density (Webber, 2003) and means that glycogen synthesis and catabolism involves alterations in fluid balance (Hall *et al.,* 2012). Carbohydrate stores are also closely regulated by adjusting oxidation to intake, making fat the substrate that is primarily used or stored in response to day-to-day fluctuations in energy balance (Abbott *et al.,* 1988). In contrast with proteins and carbohydrates, the storage capacity of fat in the form of triglycerides is virtually unlimited in the human body (Brotman & Girod, 2002). This storage is achieved without water accumulation through increases in adipocyte size and/or number (Knittle *et al.,* 1979). The ability to efficiently store triglycerides combined with the limited carbohydrate storage capacity explains why excessive intake of carbohydrates potentially leads to fat accumulation. This can either be achieved indirectly by reducing the need for fat as fuel, or directly by conversion of excess carbohydrate into fat through lipogenesis (Aarsland *et al.,* 1997). Moreover, the oxidation of fat is, contrary to carbohydrates, mainly determined by the presence or absence of other macronutrients (Melzer, 2011). Therefore, diet composition clearly has an important influence on energy balance and the regulation of body mass. For instance, a change in the

macronutrient composition of the diet can have an impact on body composition (Goss *et al.,* 2012) and energy expenditure (Ebbeling *et al.,* 2012) independent of its energy content.

2.3.2 Energy intake

Energy intake includes three major macronutrient groups (i.e. protein, carbohydrate and fat) and alcohol. After ingestion, the absorption of protein (~ 4 kcal/g, 17 kJ/g), carbohydrate (~ 4 kcal/g, 17 kJ/g), fat (~ 9 kcal/g, 38 kJ/g) and alcohol (~ 7 kcal/g, 29 kJ/g) is variable with approximately 5-10% of the ingested energy being lost in the urine and faeces (Atwater & Rosa, 1899; Norgan & Dumin, 1980). Indeed, the inter-individual variability of energy intake (i.e. absorbed or metabolised energy) is dependent on potential intestinal factors (e.g. gut microbes) and diet composition (e.g. fibre content). In a study conducted by Jumpertz and colleagues (2011) the observed associations between gut microbes and nutrient absorption suggests a possible role of the human gut microbiota in the regulation of the nutrient absorption. Previous studies have also reported that individuals who consumed high-fibre diets exhibited a higher faecal energy loss than individuals who consumed low-fibre diets with equivalent energy content (Beyer & Flynn, 1978; Wisker & Feldheim, 1990; Wisker *et al.,* 1988).

In summary, energy intake is determined by the difference between the gross energy of ingested nutrients and the energy losses found in the faeces and urine, however because of their measurement (by bomb calorimetry) often being impractical the Atwater (1899) general values for metabolised energy are used. Therefore, energy intake is often estimated from the amount of food ingested that can be assessed by self-reported techniques (e.g. food diaries with or without weighing, 24-hour dietary recall and food

frequency questionnaires) or direct observation (e.g. covert continuous weighing of participants food in a laboratory environment with instruments such as the Sussex Ingestion Pattern Monitor) (Hubei *et al*., 2006; Johnson, 2002).

2.3.3 Energy expenditure

Total energy expenditure (TEE) includes the energy used for all biological processes of the body and includes basal metabolic rate (BMR), thermic effect of food (TEF) and physical activity energy expenditure (PAEE). BMR can account for approximately 45-70% of the total daily energy expenditure (WHO, 2004) and reflects the energy needed to maintain the metabolic activities of cells and tissues and the energy needed to sustain blood circulation, respiration, and gastrointestinal and renal function (Butte *et al.*, 2012). This component of TEE varies as a function of body size, body composition and energy imbalance (Hall *et al.*, 2012) and its measurement is often made by indirect or direct calorimetry in the morning with individuals in the fasted state, resting in a still and supine position in a thermoneutral environment (Levine, 2005; Valanou *et al.*, 2006).

The TEF can account for 6-12% of total daily energy expenditure (Levine *et al.*, 2006) and is defined as the energy used for the digestion, absorption and storage of food and is primarily determined by the amount and composition of the foods consumed (Levine & Kotz, 2005). This component can be estimated through the measurement of preprandial and postprandial energy expenditures by indirect calorimetry (D'Alessio *et al.*, 1988) or, as it often is, assumed to be 10% of total daily energy expenditure in individuals consuming a mixed diet and in energy balance (Westerterp, 2001).

PAEE is the energy expended during any physical activity and can be further divided into exercise energy expenditure and non-exercise energy expenditure also known as non-exercise activity thermogenesis (NEAT) (Levine *et al.,* 1999). PAEE is the most variable and, after BMR, the second largest component of total daily energy expenditure (Ravussin & Bogardus, 1989). This variability among free-living individuals is due to their physical activity level (PAL) (Westerterp, 2008), which can be defined as 24 hours TEE expressed as a multiple of BMR, and calculated as TEE/BMR (WHO, 2004) (Table 2.1). This allows a direct comparison of activity patterns since confounding variables such as stature, body mass, sex and age have largely been removed (Livingstone *et al.,* 1991). Therefore, PAEE is able to, as a proportion of total daily energy expenditure, vary from 5% in a participant with a PAL of 1.2 to 45-50% in a participant with a PAL of 2.2-2.5 (Westerterp, 2003).

Table 2.1 Classification of lifestyles in relation to PAL in adults (from WHO, 2004)

Category	PAL value
Sedentary or light activity lifestyle	1.40-1.69
Active or moderately active lifestyle	1.70-1.99
Vigorous or vigorously active lifestyle	2.00-2.40*

* PAL values > 2.40 are difficult to maintain over a long period of time.

In addition to the values shown in table 2.1, some individuals can in extreme cases have a PAL lower than 1.40 (e.g. 1.21 in non-ambulatory adolescents with myelodysplasia) and more than 2.40 (e.g. 4.5 to 4.7 during three weeks of competitive cycling), however the former is not compatible with long-term health and the latter is difficult to maintain in the long-term (WHO, 2004). As described above, BMR is largely determined by body size and composition while the TEF is a small component of total daily energy

expenditure (6% to 12%) (Levine *et al*., 2006), so the variance in energy expenditure between individuals with similar body size can only be explained by variance in PAEE (Levine & Kotz, 2005). To achieve this, PAEE is estimated through subjective (e.g. physical activity questionnaires, 24-hour recall, and activity diaries) or objective methods (e.g. pedometer, accelerometers, heart rate monitors or multisensor devices such as the Actiheart) (Andre & Wolf, 2007).

2.3.4 Is energy intake matched to energy expenditure?

From a biological perspective it is plausible to expect energy intake to be matched to energy expenditure as the latter provides the energy needed for all metabolic and behavioural activities. Indeed, the belief that the existence of balance between energy intake and expenditure over time postulated the presence of some form of control (Dumin, 1961) that lead researchers to "*desire to find out more about the mechanisms which relate intake to expenditure - what regulates appetite, in fact"* and ask "*how quickly and closely need the supply conform to the demand?"* (Edholm *et al.* 1955, p. 297). To achieve this, Edholm and colleagues (1955) devised a study where energy intake and expenditure of twelve young male cadets in a training establishment of the armed forces were directly measured daily for a fortnight with diary and timing techniques. In this study there was a correlation between energy intake and expenditure over the 7- and 14-day period and between the mean expenditure on one day and the intake 2 days later, yet there was no correlation within the same day. Thus, this study suggested that energy intake and expenditure were not matched daily but rather on a weekly basis and that there was a 2-day lag between energy expenditure and food intake.

In a meta-analysis of six studies (69 participants in total) conducted by Dumin (1961), only 41 participants (59%) matched their energy intake and expenditure on a weekly basis and there was no evidence supporting the idea of the 2-day lag between energy expenditure and food intake. Dumin suggested that these results made unlikely the presence of any day-to-day control of energy balance and that appetite was more likely controlled by a double mechanism, *"(1) a gross, short-term regulation and (2) a long-term one, operating with a delay of days, weeks or occasionally months*" (Dumin, 1961, p. 303). However, Dumin also recognised that despite the unlikelihood of the presence of fine-tuned control mechanisms responsible to attain daily energy balance, it was possible that one may exist to control food intake in the short-term specially in the presence of exercise-induced high energy deficits (Dumin, 1961). For instance, other studies have observed the ability to maintain short-term energy balance in 5 cyclists during the Tour de France that matched their energy intake and expenditure over a period of 3 days (Saris *et al.,* 1989) and 8 Swedish national team cross-country skiers that matched their energy intake and expenditure during pre-season training camp over a 6- (men) and 7-day (women) period (Sjodin *et al.,* 1994). Therefore, it is possible that the mechanisms responsible for matching energy intake and expenditure over the short-term may be improved in well-trained individuals.

Another study by Edholm and colleagues also failed to observe the 2-day lag effect but the authors suggested that this result was not because of a lack of presence of a lag between expenditure and intake but rather that the duration of the lag was variable both between- and within- individuals (Edholm *et al.,* 1970). Additionally, these findings illustrated the difficulty of establishing the relationship between energy intake and expenditure taking into consideration the limited accuracy with which these components

can be measured and used to calculate energy balance (Edholm, 1973; Edholm *et al.*, 1970). The measurement of daily energy expenditure was of special concern, as it is very difficult to achieve without interfering or disturbing the individuals' daily routine (Edholm, 1973). In an attempt to overcome this limitation, De Castro (1997) examined how individuals could achieve energy balance in the free-living using pocket-sized dietary records and triaxial accelerometers during a 7-day period. No correlations were observed between total daily activity and total intake on the same day or any of the subsequent four days. After performing autocorrelations for energy intake and activity over the 7-day period, De Castro (1997) reported that intake and activity on one day had a maximum negative effect on energy intake and activity two to three days later. However, the sizes of the effects were small with the largest intake autocorrelation accounting for less than 4% of the variance in daily intake and the largest activity autocorrelation accounting for only 6% of the variance in daily activity.

In summary, most of these studies support that there is a weak relationship between energy intake and expenditure in the short-term (1-2 days) and that this relationship is improved over a longer period of time (e.g. 7 days), yet these studies (De Castro, 1997; Dumin, 1961; Edholm *et al.*, 1970; Edholm *et al.*, 1955) were observational in nature which, despite allowing the determination of a relationship between the variables, does not allow inferences on causality. To overcome this limitation, several experimental studies (Hubert *et al.*, 1998; Stubbs *et al.*, 2004a; Stubbs *et al.*, 2004b) have examined the relationship between the main behavioural determinants of energy intake (i.e. eating) and expenditure (i.e. physical activity). Therefore, the next sections of this literature review will focus on the control of appetite, physical activity and their relationship.

2.4 Appetite

An understanding of the complex psychobiological system of appetite control is vital to energy balance (Benelam, 2009), therefore this section will focus on defining some terms that are often used in this area (i.e. appetite, hunger, satiation and satiety) and presenting the physiological, behavioural/psychological and environmental factors that influence appetite.

2.4.1 Definitions of appetite, hunger, satiation and satiety

According to Blundell *et al.* (2010) there are currently two definitions of the term appetite, so to avoid confusion appetite will be referred throughout this thesis as a general term which covers the field of food intake, sensation, motivation and preference. Hunger is defined as a conscious sensation reflecting a mental urge to eat that can be accompanied by feelings of light headedness, weakness or emptiness in the stomach (Blundell *et al.*, 2010). Regarding satiation and satiety, the first is defined as the process that leads to the termination of eating (i.e. determines how much food is eaten and the duration of food intake) while the latter is the process that leads to inhibition of further eating (i.e. determines when food is eaten and the time between meals) (Benelam, 2009).

2.4.2 Physiological factors

The physiological control of appetite is mediated by a highly complex system involving two complementary drives; the homeostatic pathway which focuses primarily on the maintenance of energy balance, and the hedonic pathway, which involves the rewarding value of food (Lutter & Nestler, 2009).

2.4.2.1 Homeostatic pathway

The homeostatic pathway involves short-term and long-term neural and hormonal signals which give the brain information regarding the body's nutritional status (Murphy & Bloom, 2006). Short-term (i.e. episodic) signals are triggered by eating episodes and originate mainly from the gastrointestinal tract (Blundell, 2006). These signals involve mechanical (e.g. gastric distension) and chemical components (e.g. cholecystokinin (CCK), oxyntomodulin, peptide tyrosin-tyrosin (PYY), glucagon-like peptide 1 (GLP-1) and ghrelin) that contribute to hunger, satiation and/or satiety (Berthoud, 2008). For example, CCK and oxyntomodulin provide signals leading to meal termination (i.e. satiation), while others such as GLP-1 and PYY are also involved in inhibiting further eating during the postprandial period (i.e. satiety) (Blundell *et al.,* 2008). Unlike these short-term peptides ghrelin is the only known orexigenic (appetite-stimulating) gut hormone suggested to have a role in meal initiation due to its preprandial rise and postprandial fall (Cummings *et al.,* 2001) and ability to induce an increase in food intake after intravenous infusion (Wren *et al.,* 2001). Conversely, the long-term (i.e. tonic) signals (e.g. leptin and insulin) are associated with the metabolic state of adipose tissue and secreted from tissue stores (Blundell, 2006).

One of the most important long-term signals is leptin which is produced by adipose cells in proportion to body fat mass and signals the level of energy stores to hypothalamic receptors that control energy intake and expenditure (Jequier, 2002). Insulin, a peptide secreted by the beta-cells of the islets of Langerhans in the pancreas, is another important long-term signal that circulates in proportion to body fat stores and induces a long-term catabolic response, decreasing food intake and increasing energy expenditure (Konner *et al.,* 2009). Leptin and insulin receptors are widely expressed

within the brain, including an important site for the homeostatic control called arcuate nucleus of the mediobasal hypothalamus. This area of the hypothalamus contains two functionally opposing sets of neurons: the orexigenic (appetite stimulating) agouti-related peptide/neuropeptide Y (AgRP/NPY) and the anorexigenic (appetite suppressing) proopiomelanocortin/cocaine and amphetamine regulated transcript (POMC/CART) (Garfield *et al*., 2009) which respond to changes in short- and long-term metabolic signals (Figure 2.1). The AgRP/NPY are inhibited by leptin, insulin and PYY and stimulated by ghrelin, whereas the POMC/CART are stimulated by leptin and inhibited by neighbouring AgRP/NPY neurons (Morton et al., 2006). Additionally, the POMC neurons are able to produce alpha-melanocyte stimulating hormone (a-MSH), an anorectic peptide that acts on melanocortin receptors types 3 and 4 to reduce food intake and energy expenditure in a manner similar to leptin (Boston *et al.,* 1997). Therefore, the homeostatic pathway includes a complex array of signals which can act via circulating hormones (e.g. ghrelin) or neural connections (e.g. vagus nerve). The integration of these signals allows the brain to recognise the current dynamic state of energy stores (long-term signals) and the flux of ingested nutrients (short-term signals) which are used to induce appropriate compensatory metabolic responses (Berthoud, 2008).

Figure 2.1 Representation of short- and long-term signals influencing energy homeostasis via the arcuate nucleus (adapted from Murphy & Bloom 2004). Continuous lines indicate stimulatory effects, and dashed lines indicate inhibitory effects. AgRP, agouti-related peptide; CART, cocaine and amphetamine regulated transcript; GLP-1, glucagon-like-peptide 1; aMSH, alpha-melanocyte-stimulating hormone; NPY, neuropeptide Y, POMC, proopiomelanocortin; PYY, peptide YY.

2.4.2.2 Hedonic pathway

Eating is essential for survival, therefore it is logical that this behaviour is subject to homeostatic controls described in the previous section. However, this behaviour is also driven by factors beyond the vital need for energy such as the pleasure induced by the sensory properties of food (e.g. taste, smell and texture) which can promote eating even in the presence of physiological satiety signals (Mela, 2006; Rolls, 2012). This effect is mediated by the hedonic pathway which similarly to the homeostatic process, involves several feedback and neural systems.

The endogenous dopaminergic and opioid systems have been for a long time considered the most important systems in mediating the processes of brain reward. However, recent evidence suggests that the endogenous cannabinoid (endocannabinoid) system has also an important role in the signalling of rewarding events (Solinas *et al.,* 2008). The information acquired through these systems is then primarily processed in a part of the brain called the nucleus accumbens, where the dopamine, opioids and endocannabinoids signals interact to modulate responses to food (Fulton, 2010). Food consumption increases brain dopamine levels in humans (Small *et al.,* 2003) which when altered can reduce (dopamine agonists) or increase (dopamine antagonists) energy intake (Leddy *et al.,* 2004; Wellman, 2005). The role of dopamine in the hedonic processing is also supported by positron emission tomography scans, indicating that an increase in dopamine release is correlated with the degree of experienced pleasure (Blundell, 2006). The opioids can act in the nucleus accumbens to increase food reward and stimulate food intake (Pecina, 2008). Moreover, opioids influence the dopamine reward system by inhibiting GABAergic input to the dopamine neurones of the ventral tegmental area (VTA), resulting in increased dopamine release (Spanagel & Weiss, 1999). The endocannabinoids control the rewarding effects of food by the activation (enhances food reward) or deactivation (diminishes food reward) of cannabinoid type 1 receptors (Di Marzo & Matias, 2005). These receptors are present in the VTA where they increase dopamine firing and release (Cheer *et al.,* 2004). Recent evidence also suggests that the endocannabinoid system is able to influence the homeostatic pathway by controlling food intake via both central and peripheral mechanisms and possibly stimulate lipogenesis and fat acumulation (Di Marzo & Matias, 2005). In addition to this interaction between dopamine, endocannabinoids and opioids in the control of food reward there are several homeostatic metabolic signals (e.g. leptin, insulin and ghrelin)

that affect the hedonic pathway (Figlewicz & Benoit, 2009). There are receptors for these hormones on dopamine neurones in the VTA, to which ligand binding results in activation by ghrelin (Abizaid et al., 2006) or inhibition by leptin and/or insulin of dopamine signalling to the nucleus accumbens (Magni et ah, 2009). These alterations in dopamine signalling pathways have complex effects on eating behaviours and can possibly explain why imaging studies show that obese participants can have impairments in dopamine food reward pathways (Volkow *et al*., 2011).

In summary, the hypothalamus integrates various signals to control appetite with a homeostatic purpose of regulating body mass. Additional neural circuits have the ability to override these homeostatic signals, which may result in either gluttony or anorexia at the extremes (Kaye *et al*., 2013; Palmiter, 2007). Despite being primarily controlled by different areas of the brain (the homeostatic process being associated primarily with the hypothalamus and brainstem and the hedonic process with the limbic regions and cerebral cortex) (Ahima & Antwi, 2008), recent evidence suggests that both pathways are intimately linked by internal metabolic signals (Berthoud & Morrison, 2008). Still, uncertainty remains whether they operate independently or interact with each other (Berthoud, 2011; Saper *et al.,* 2002).

2.4.3 Behavioural/psychological factors

In addition to the physiological mechanisms there are several behavioural/psychological factors that can influence food intake. These include expected satiety and familiarity with foods (Irvine *et al.,* 2012), eating habits (Gardner et al., 2011), acquired food preferences (Teixeira *et al.,* 2011), intentions to eat (Adriaanse *et al.,* 2011), attitudes towards food (Babicz-Zielinska, 2006), emotional states (Geliebter & Aversa, 2003),

stress (Groesz *et al.,* 2012) and other eating behaviour traits (Lowe *et al.,* 2009). In particular, the factors of cognitive restraint and disinhibition have emerged as important eating behaviour traits that influence body mass and therefore are deemed as a psychological marker of appetite control (King *et al.,* 2012). For example, many individuals eat whatever they want whenever they want while others attempt to assert cognitive control over their food intake. The latter process, referred to as cognitive restraint (De Castro, 1995), is used to describe the type of behaviour where someone exerts cognitive control to limit food intake for the purpose of maintaining or losing body mass (Herman & Polivy, 2005). Conversely, disinhibition can be described as the loss of cognitive restraint that confers a susceptibility to an increase in body mass (Rogers, 1999) or as suggested more recently 'opportunistic eating' (Bryant *et al.,* 2007). High levels of cognitive restraint have been associated with intent to diet and controlled eating (Lawson *et al.,* 1995), however, this relationship may be disrupted by events related to the disinhibition of cognitive control of eating behaviour leading to episodes of overeating (King, 1999). For instance, when focusing on food or a low cognitive load restrained eaters are able to limit their intakes at meal times, but distraction or high cognitive loads increases intake especially in those individuals with higher restraint scores (Bellisle & Dalix, 2001; Mann & Ward, 2004). Therefore, when restrained eaters are faced with cues that remind them of their intention to restrict food intake restraint is increased, but if attention is narrowed to other cues then this situation promotes disinhibited eating (Mann & Ward, 2007). For example, listening to music or watching television while eating is associated with an increase in energy intake (Bellisle *et al.,* 2004; Stroebele & De Castro, 2006). Indeed, high levels of disinhibition have been shown to be strongly associated with an increase in body mass over time and obesity in adult life (Provencher *et al.,* 2003). Nevertheless, it is important to consider

that other individual differences in the response to situational cues involving food may also play a role in susceptibility to overeating and increase of body mass in the long term (Hetherington, 2007).

2.4.4 Environmental factors

There is clear evidence that appetite can be controlled via several physiological and behavioural/psychological factors, however, it is important to consider that these are influenced by the environment. The latter can be divided into physical (what is available), economic (what are the costs), political (what are the rules), and sociocultural (what is the social and cultural background) environments (Kremers *et al*., 2006). The physical environment includes factors such as the types of foods available in the residential area (Morland *et al.,* 2006), schools (Kubik *et al.,* 2003) or workplaces (Booth *et al.,* 2001) which nowadays tend to include "super-size" portions of a variety of highly palatable energy dense foods (Hill & Peters, 1998; Smith & Ditschun, 2009). Other factors include package size, variety, plate size (Wansink, 2004) and ambient characteristics such as lightning, temperature and altitude (Westerterp-Platenga, 1999). In addition, the physical environment may also trigger eating habits. For instance, in a recent study Neal and colleagues (2011) reported that university students' ate more popcorn in the cinema context as compared with the meeting room context, supporting the hypothesis that environmental cues trigger habitual eating behaviour. The economic environment includes the cost of food and economic status which can play an important role in determining eating patterns (e.g. reduced consumption of fruits, vegetables and dairy products by lower income families due to economic constraints) (Darmon *et al.,* 2003; Darmon & Drewnowski, 2008; Kirkpatrick & Tarasuk, 2003; Monsivais & Drewnowski, 2007). The influence of the political environment (i.e. policies and laws)

on eating behaviour is mostly determined by its relation with some of the factors presented above (e.g. it can influence the location of fast food restaurants and food prices) (Epstein *et al*., 2010; Neal *et al*., 2006; Story *et al.,* 2008). In contrast, the social and cultural environment has been shown to directly influence food intake by factors such as the presence of others during a meal (Herman & Polivy, 2005) or cultural norms around body size and food (Swinbum *et al.,* 2011). Studies based on the food-diary method have shown that meal size is influenced by the number of persons at the meal (De Castro, 1995) and that meals could be increased up to 75% in large groups compared to when alone (De Castro & Brewer, 1992). However, this is not always the case as recent evidence support that social influence may induce inhibitory and stimulatory effects on intake. This observed effect is possibly mediated by several factors such as the duration of the meal, the gender and familiarity of guests sharing the meal, and the actual circumstances of the meal (Bellisle *et al.,* 2009). An increased amount of food may be eaten at meals with familiar and friendly people whereas it may be decreased when eating in the presence of unfamiliar people. In the former the familiarity may help to make a meal relaxing and more enjoyable reducing an individual's ability or motivation to monitor consumption while in the latter food the presence of an unfamiliar person may increase the individual self-monitoring and self-awareness (Wansink, 2004). Herman and Polivy (2005) have also suggested that in the presence of others, people use others food intake to determine how much is acceptable for them to eat.

2.5 Physical activity

Physical activity improves health and promotes different physiological changes (Powell *et al.,* 2011). These changes can occur during and in the hours/days after one bout of exercise (i.e. acute effects) or occur over time due to regular exercise (i.e. chronic effects). Despite this clear difference, the acute response to an exercise bout and the chronic adaptations to exercise training cannot be considered in isolation because frequent repetition of single exercise bouts with transient responses produces permanent adaptations (i.e., chronic effect) (Kesaniemi *et al.,* 2010). In addition, physical activity is the most variable component of total daily energy expenditure (Ravussin & Bogardus, 1989; Westerterp, 2008) making it an ideal target to affect energy balance and the regulation of body mass. Therefore, to improve understanding of this behaviour this section will present various physiological, behavioural/psychological and environmental factors that influence physical activity.

2.5.1 Physiological factors

The control of physical activity has mainly been considered as voluntary and/or influenced solely by environmental factors, however, current research points to a strong biological input with potential genetic, neurological and endocrinological origins (Bowen *et al.,* 2011; Lauderdale *et al.,* 1997; Joosen *et al.,* 2005; Perusse *et al.,* 1989; Simonen *et al.,* 2003; Stubbe *et al.,* 2006). This evidence may include an increase or decrease of physiological signals, and/or genetics that may modify receptor/protein interaction (Leamy *et al.,* 2008; Lightfoot *et al.,* 2004; Lightfoot *et al.,* 2008; Mustelin *et al.,* 2012; Stubbe et al., 2005). Recent reviews have suggested a role for sex hormones (e.g. oestrogen, progesterone and testosterone) (Lightfoot, 2008) and other

biological factors (e.g. agouti-related protein, ghrelin, neuropeptide Y, corticotrophin releasing hormone, cholecystokinin, leptin, and dopamine) (Kotz *et al.,* 2008) in the control of physical activity, however this evidence is derived mostly from animal research. In humans, the neurobiological aspects of physical activity control by the central nervous system are still not understood with most research focussing in understanding central fatigue (i.e. a reduction in the drive to motor neurons) during prolonged exercise in humans (Butler *et al.,* 2007; Nybo, 2007; Rasmussen *et al.,* 2006; Ross *et al.,* 2007). Conversely, the study of the neurobiological control of non-strenuous physical activity in humans has received less attention, possibly due to the necessary ethical limitations of experimental manipulations (Bowen et al., 2011; Dishman *et al.,* 2006). Therefore, based on the vast animal research and limited human research there is evidence suggesting a biological control of physical activity (Garland *et al.,* 2011), however, uncertainty remains regarding how this control is exerted and what mechanisms are involved (Figure 2.2).

Figure 2.2 Hypothetical representation of the biological regulation resulting in differential controls of physical activity in male and female humans. Double headed arrows indicate potential two-way influences. GLUT-4, glucose transporter type 4; NPY, neuropeptide Y; ? indicates effects that are uncertain due to minimal or conflicting research (adapted from Bowen *et al.,* 2011).

2.5.2 Behavioural/psychological factors

Similarly to appetite, physical activity is a behaviour influenced by many behavioural/psychological factors. These can include lifestyle habits, beliefs/attitudes towards physical activity, emotional state, motivation, expectations (Bauman *et al.,* 2002; Gardner *et al.,* 2011), self-image, self-efficacy (i.e. one's confidence in its ability to take the steps necessary to be regularly physically active) (Anderson *et al.,* 2006; Koeneman *et al.,* 2011) and personality traits (Rhodes & Smith, 2006). For instance,

adults with a history of participation in sports in their youth have been found more likely to be physically active, and engage in vigorous activities than people who did not engage in sports in their youth (Dishman *et al.,* 1985). More recently, the subjective perception of the exercise experience has also been suggested to influence mood which in turn relates to intentions and motivation to exercise in the future (Bryan *et al.,* 2007).

2.5.3 Environmental/social factors

Physical activity is strongly influenced by environmental/social factors. For instance, the advances in technology and transportation have reduced the need for physical activity in daily life (Hill & Peters, 1998). Aspects of the home environment such as the presence of televisions and electronic games or exercise machines have been associated with changes in physical activity behaviour (Booth *et al.,* 2001). Other factors, such as the proximity to destination, travel costs and environment quality (e.g. good outdoor facilities such as access to green spaces) are likely to be influential (Frank *et al.,* 2005; Haug *et al.,* 2010; Saelens *et al.,* 2003). The socioeconomic environment composed of economic factors (e.g. poverty rate) and social factors (e.g. racial composition or crime and safety), is also associated with physical activity (Lovasi *et al.,* 2009; Wen *et al.,* 2007). Additionally, the influence of a spouse's behaviour and social support (i.e. the perceived support for physical activity received from others, such as family and friends) can also have an effect on physical activity behaviour (Anderson *et al.,* 2006; Gorin et al., 2008).

2.6 Relationship between physical activity and appetite

Several decades ago it was purported that "*the regulation of food intake functions with such flexibility that an increase in energy output due to exercise is automatically followed by an equivalent increase in caloric intake*" (Mayer *et al.* 1956, p. 169). To investigate this relationship, Mayer and his colleagues (1956) compared energy intake with the physical demand of the occupation of 213 men employed in a jute factory in India. Energy intake was measured by dietary interviews using pre-established questionnaires based on cross checks with results being analysed using the Indian Food Composition Table (Mayer *et al.* 1956). The individuals' occupational activity status was assessed by a combination of detailed observation of the physical demand of each work-based task undertaken, a thorough analysis conducted by an efficiency engineers' firm and the use of indirect calorimetry data for occupational categories in the jute industry. This allowed the classification of the occupations in order of increasing energy cost into five categories: first class (i.e. sedentary), second class (i.e. light work), third class (i.e. medium work), fourth class (i.e. heavy work) and fifth class (i.e. very heavy work) (Mayer *et al.* 1956). In this study, energy intake increased with activity only within certain zones ("normal activity range") and below that range ("sedentary zone") a decrease in activity was not followed by a decrease in energy intake but rather an increase (Figure 2.3). Therefore, these results suggest that energy compensation may exist within the "normal activity range" and that the ability to control energy intake is possibly dependent on the level of habitual physical activity. Additionally, body mass was increased in the first class group (i.e. sedentary) lending support to a possible inability of these individuals to regulate energy balance.

Figure 2.3 Diagram from Mayer *et al.* **(1956)** that illustrates the relationship between physical work and food intake with the addition of the "sedentary" and "normal activity range" zones for easier interpretation. Circles refer to the mean intake per occupation (e.g. supervisors) and crosses to the mean intake per category (e.g. sedentary).

Mayer and colleagues (1956) study was observational which does not allow any inferences regarding cause and effect. Additionally, the usual classification of energy requirements by occupation alone may give a poor prediction of total energy expenditure (Livingstone *et al.,* 1991). To overcome this latter limitation, Schoeller (1998) examined Mayer's hypothesis (i.e. that the mechanisms controlling energy balance are accurate in individuals with high levels of physical activity but not in sedentary individuals) by reviewing cross-sectional and longitudinal data from doubly labelled water studies in adults. Schoeller (1998) suggested that these studies supported Mayer's hypothesis in men but not in women, suggesting a possible sex difference in the regulation of body mass in the response to physical activity.

Studies have also investigated the relationship between individuals' physical activity and the ability to control energy intake in the short-term using the preload-test meal paradigm (King *et al.,* 1999; Long *et al.,* 2002; Van Walleghen *et al.,* 2007). These studies manipulated the energy content of a preload and measured energy intake at subsequent test meals. King and colleagues (1999) suggested that active men were able to compensate for the energy content of the post-exercise preload drink, by adjusting energy intake at the subsequent meal. Long and colleagues (2002) compared this effect in male regular exercisers and non-exercisers, and suggested that regular exercisers had an increased accuracy of short-term control of food intake compared with non-exercisers. In this study, regular exercisers were able to compensate for the preload manipulation by reducing energy intake at the buffet meal after the high-energy preload compared to the low-energy preload. In contrast, non-exercisers were not able to compensate for the energy difference between the preloads and had approximately the same energy intake at the buffet lunch on both conditions. Similarly, another study investigated this relationship in active and inactive, young and old adults using a no-preload versus preload condition and measuring food intake at both a buffet meal and over the course of the testing day (Van Walleghen *et al.,* 2007). This study, reported that the ability to acutely (i.e. at the buffet lunch meal) control energy intake was influenced by age, with young adults compensating better than older adults, but not by habitual physical activity. Conversely, the short-term energy control (i.e. over the course of the testing day) was more accurate in active compared to inactive adults, independent of the age supporting the beneficial role of exercise on the short-term control of appetite. Davy *et al.* (2007) investigated possible sex differences in appetite control by comparing energy intake compensation after a preload or no preload in 12 men and 12 women matched for age, habitual physical activity, cardiorespiratory

fitness, and dietary cognitive restraint. In this study, participants were given either a yogurt preload (representing -26 kJ/kg body mass for both men and women) or no preload thirty minutes before the lunch meals. Energy intake compensation for the yogurt preload was more accurate in men than women (86.2 ± 5.0 vs. 73.6 ± 4.8% compensation) but there were no differences in subjective hunger ratings. Findings suggest that using acute test meals, men control their energy intake more accurately than women, which if persistent may predispose women to increases of body mass over time (Davy *et al*., 2007).

Despite being experimental, inferences made from these studies are limited due to their cross-sectional design as the observed effects may be due to lifestyle or other factors and not the individuals' physical activity. Recent intervention studies (Martins *et al.,* 2013; Martins *et al.,* 2007b) have addressed this limitation by examining the chronic effects of exercise on energy compensation in response to a high- and low-energy preload in inactive adults. In one study, energy intake at a buffet lunch meal and subsequent 24 hours was measured at baseline and after 6 weeks of non-supervised exercise sessions (4 times per week, 65-75% maximal heart rate) (Martins *et al.,* 2007b). This study reported no differences in the buffet energy intake after two preloads at baseline for both men and women and a lower buffet energy intake after the high-energy preload compared to the low-energy preload following the exercise intervention only in men. When analysing the cumulative energy intake there were no differences between preloads at baseline and after the 6-week exercise intervention in both men and women (Martins *et al.,* 2007b). In another study, energy intake at an *ad libitum* test meal (pasta with tomato sauce and cheese) and over the subsequent 24 hours was measured at baseline and after 12 weeks of supervised exercise sessions (5 days a week,

75% maximal heart rate) (Martins *et al.,* 2013). In this study, there were no differences in the test meal energy intake after the two preloads at baseline and after 12 weeks. However, cumulative energy intake after the high-energy preload was higher than after the low-energy preload at baseline, while after the 12-week exercise programme the opposite was observed (i.e. cumulative energy intake after the high-energy preload was lower than after the low-energy preload) (Martins *et al.,* 2013). Results from these longitudinal studies support previous cross-sectional findings that exercise may have a beneficial role in the short-term control of appetite and that this leads to a more sensitive response to previous energy intake. Moreover, findings from one of these studies (Martins *et al.,* 2007b), support a possible sex difference in the short-term control of appetite after an exercise intervention which may possibly be associated with the previous observation by Schoeller (1998) of a possible physical activity related sex difference in the regulation of body mass.

Notwithstanding the evidence that studies using the preload-test meal paradigm can provide regarding the relationship between physical activity status and the ability to control energy intake in the short-term, their findings are specific to situations where energy intake is manipulated. Therefore, Hubert *et al.* (1998) investigated the effects of two methods of inducing an acute energy deficit (by exercise and a low-energy breakfast) on appetite. This study reported that energy intake at lunch was increased after the low-energy breakfast but not after exercise suggesting that the body may respond differently to an energy deficit created by reducing energy intake and increasing exercise, respectively. A recent study (King *et al.,* 2011) has strengthened the support for these reported differences as the appetite and *ad libitum* energy intake of 12 healthy men changed in a compensatory fashion to food restriction but not to exercise.

In contrast with these studies, Murgatroyd and colleagues (1999) investigated the effects of an acute energy surplus (inactivity) and dietary macronutrient composition (containing either 35% or 60% energy as fat) on energy balance in 8 men. This study reported no changes in energy intake over a 2-day period in response to either the imposition of inactivity or a high-fat diet. The authors suggested a possible adverse role for inactivity and a high-fat diet, and a beneficial role for physical activity, on the regulation of energy balance (Murgatroyd *et al*., 1999). These findings were further supported by Shepard *et al.* (2001) that examined the effect of the interaction of diet composition (high-fat or high-carbohydrate) and physical inactivity on energy and fat balances. After consuming the high-fat diet and remaining physical inactive -80% of all individuals were in positive fat balance whereas after the high-carbohydrate diet with physical inactivity only -50% of participants were in positive fat balance (Shepard *et al.,* 2001). However, the measurement period in these previous studies was short in duration (< 2 days), a limitation which was later addressed by a study examining how a seven day regime of inactivity affected appetite within a whole-body indirect calorimeter (Stubbs *et al.,* 2004a). This study reported no compensatory reduction of energy intake suggesting that chronic inactivity may impair the ability to regulate energy balance, leading to a state of positive energy balance which over time would result in the accumulation of body fat (Stubbs *et al.,* 2004a).

In summary, the observed acute or short-term adjustment of food intake in response to previous energy intake supports a beneficial role for physical activity on energy intake and possible sex differences in the short-term control of appetite. There is also evidence that individuals respond differently to manipulations in energy intake and energy expenditure. To better understand the relationship between physical activity and

appetite there is a need to investigate the effects of exercise-induced energy deficits on energy intake. Moreover, it is important to recognise the acute and chronic effects of physical activity interventions on eating behaviour as these may have different implications for body mass management strategies. The following sections will review the current evidence from experimental studies that investigated the effects of physical activity on appetite. Since physical activity in these studies is prescribed the term "exercise" will be used throughout.

2.6.1 Exercise, hunger and energy intake

Exercise is a recognised obesity prevention and management strategy but the extent to which it beneficially affects appetite control is not clear (Martins *et al.,* 2008). The efficacy of exercise in the prevention of increases in body mass and promotion of reductions in body mass varies between individuals so there is a need to understand if possible behavioural and physiological compensatory responses account for these differences (King *et al.,* 2007) and if these compensatory responses differ according to participants' habitual physical activity (Martins *et al.,* 2008) and sex (Hagobian *et al.,* 2010). The following is a review of the current knowledge of the acute and chronic effects of exercise on hunger and energy intake in men and women.

2.6.1.1 Effects of acute exercise in men

Several studies have investigated the effects of an acute bout of exercise on food intake and hunger in men, however, controversy remains. Contrary to popular belief, hunger does not change (Imbeault *et al.,* 1997; Jokisch *et al.,* 2012; King *et al.,* 1997; King *et al.,* 2010b) or transiently decreases (Broom *et al.,* 2009; Broom et al., 2007; King *et al.,*

2010a; King *et al.,* 2013) during and after an acute bout of exercise. The latter phenomenon has been termed "exercise-induced anorexia" and is believed to be dependent on the intensity of the exercise bout (Martins *et al.,* 2008). This is supported by previous research that reported suppression of hunger after high-intensity (68-70% of V02max) but not after low-intensity (30-35% of 35% of V02max) bouts of exercise eliciting similar energy deficits (King et al., 1994; Thompson *et al.,* 1988). Although studies have reported a decrease in hunger with moderate intensity exercise bouts (-50% of V02max) (Ueda *et al.,* 2009b; Vantansever-Ozen *et al.,* 2011), it is generally accepted that the exercise-induced anorexia phenomenon tends to only occur with vigorous exercise (>60% of V02max) (Bilski *et al.,* 2009; Blundell & King, 2000; Martins *et al.,* 2008). Additionally, this exercise-induced hunger suppression is often short-lived lasting from 15 minutes (King & Blundell, 1995; King *et al.,* 1994) up to two hour after the end of the exercise (Broom *et al.,* 2009; Broom *et al.,* 2007), making it unlikely to have any significant impact on subsequent energy intake.

The transient suppression of hunger is not associated with changes in energy intake as several studies reported no effects of an acute bout of exercise on subsequent energy intake (Deighton *et al.,* 2012; Hagobian *et al.,* 2012; Kelly *et al.,* 2012; King & Blundell, 1995; King *et al.,* 1994; King *et al.,* 1997; King *et al.,* 2010a; Thompson *et al.,* 1988; Vantansever-Ozen *et al.,* 2011). Fewer studies have reported decreases (Westerterp-Plantenga *et al.,* 1997, Jokisch *et al.,* 2012; Ueda *et al.,* 2009a; Ueda *et al.,* 2009b) and increases in energy intake after an acute bout of exercise (Martins *et al.,* 2007a, Shorten *et al.,* 2009; Verger *et al.,* 1994).

King *et al.* (1994) suggest exercise-induced energy expenditure should be considered when interpreting the effects of exercise on energy intake. This notion was termed "relative energy intake" and is important because even if exercise induces an increase in energy intake this does not necessarily translate to positive energy balance (King *et al.,* 1994). Therefore, a reduction in relative energy intake indicates that a short-term negative energy balance can be attained. For instance, after accounting for the energy expended during the bout of exercise, previous studies reporting no changes (Deighton *et al.,* 2012; Kelly *et al.,* 2012) and an increase in absolute energy intake (Martins *et al.,* 2007a), showed a decrease in relative energy intake supporting that after an acute bout of exercise participants were actually in energy deficit.

Despite the need to investigate effects of exercise on food intake in the inactive, overweight and obese most studies have examined only regular exercisers (Balaguera-Cortes *et al.,* 2011; Deighton *et al.,* 2012; Imbeault *et al.,* 1997; Kelly *et al.,* 2012; King & Blundell, 1995; King *et al.,* 1994; King *et al.,* 2010a; King *et al.,* 2010b; Shorten *et al.,* 2009; Vantansever-Ozen *et al.,* 2011; Verger *et al.,* 1994). From the few studies that examined inactive-to-moderately-active men (Gilsenan *et al.,* 1998; Harris & George, 2008; Jokisch *et al.,* 2012; Klausen *et al.,* 1999; Ueda *et al.,* 2009b; Westerterp-Plantenga *et al.,* 1997) there are mixed findings with some studies reporting no change (Gilsenan *et al.,* 1998; Harris & George, 2008, Klausen *et al.,* 1999), and others a decrease in food intake after exercise (Jokisch, *et al.,* 2012; Ueda *et al.,* 2009b; Westerterp-Plantenga *et al.,* 1997). Additionally, the only study that observed an exercise-induced increase on energy intake in inactive-to-moderately-active individuals (Martins *et al.,* 2007a) included six men and six women, which limits interpretation of findings considering that sex differences may exist.

The effects of three 40 min cycling bouts (75W) on total daily energy intake were investigated in a whole-body calorimeter in habitually sedentary male participants (Gilsenan *et al.,* 1998). Despite an increase in energy expenditure of 2MJ, energy intake remained unchanged during the whole experimental day. Klausen and colleagues (1999) investigated the effects of high- (30 min at 60% of V02max) and low-intensity (60 min at 30% of V02max) cycling on the following day's energy intake in non-athletes with a healthy BMI. In this study, energy intake did not change between conditions or in comparison with habitual energy intake suggesting that contrary to hunger, the acute control of energy intake may not be dependent on exercise intensity. This is supported by previous studies in active individuals that reported energy intake to be unaffected after low- and high-intensity exercise (with similar energy deficits) on a cycle-ergometer (King *et al.,* 1994; Thompson *et al.,* 1988) and treadmill (Imbeault *et al.,* 1997). A later study by Ueda *et al.,* (2009a) comparing 30 min of moderate- (50% of V 02max) and high-intensity cycling (75% of V02max) has also reported no differences in energy intake between exercise intensities. Additionally, the duration (King *et al.,* 1994) and mode of exercise (running vs. cycling) (King & Blundell, 1995) does not appear to affect the energy intake response to intense exercise, which suggests that energy intake might be mainly dependent on the energy cost of exercise and not on its characteristics (i.e. intensity, duration and mode).

Regardless of BMI (healthy vs. overweight), energy intake has been reported to remain unchanged at the meal immediately after 60 min of moderate-intensity exercise (walking at 60-65% of maximum heart rate) and during the 12 subsequent hours (Harris & George, 2008). As energy expenditure was not measured in this study, it is not possible to determine the exercise-induced energy deficit which as pointed out by King

et al. (1994) limits interpretation of the findings. Nevertheless, these findings suggest that there is no acute effect of exercise on energy intake at the meal immediately after exercise, a finding supported by most studies with active individuals (Deighton *et al.,* 2012; Kelly *et al.,* 2012; King *et al.,* 2010a; Vantansever-Ozen *et al.,* 2011). In contrast, energy intake in 10 obese and 10 non-obese participants decreased at a meal given 10 min after 2 hours of cycling at 60% of their maximal power output (Westerterp-Plantenga *et al.,* 1997). A more recent study by Ueda and colleagues (2009b) reported that energy intake in both obese and healthy control individuals was decreased at a meal given 1 hour after the end of 60 min of moderate-intensity cycling (50% of VC^max)- Although there were no differences in the direction of how both groups acutely controlled their energy intake, the exercise-induced suppression of energy intake in this study was larger in the obese than in the healthy control group suggesting that only the intensity and not the direction of the change in energy intake might be dependent on body mass. Other possible explanations for the discrepancy in these findings are the differences in the exercise-induced energy deficits and participants habitual physical activity. For instance in the Ueda *et al.* (2009b) study the mean exercise-induced energy deficit in the obese group (2382 kJ) was higher than the one in the healthy control group (1825 kJ). The lack of an effect in Harris & George (2008) study could also be due to a smaller exercise-induced energy deficit, however, comparisons are not possible as they did not measure energy expenditure.

To the author's knowledge, only one study has directly compared the effects of an acute bout of moderate-intensity exercise (45 min of cycling at 65-75% of maximum heart rate) in active and inactive individuals of normal body mass (Jokisch *et al.* 2012). In this study, energy intake was measured at an *ad libitum* lunch buffet one hour after the end

of the exercise/resting periods and for the rest of the experimental days. Inactive participants reduced their energy intake in the *ad libitum* meal compared to control whilst there were no differences in active participants' energy intake at the same meal. When analysing the remainder of the experimental day both groups showed an increase in energy intake after exercise compared to control (Jokisch *et al.* 2012). Moreover, this study presented the percentage of energy compensation which was calculated by dividing the difference between energy intake in the exercise and control conditions by the exercise-induced energy deficit. At the *ad libitum* meal active participants demonstrated some compensation (22.7 ± 52.5%) and inactive demonstrated negative compensation (-35.5 ± 80.4%). However, when compensation was determined using the combined energy intake from the *ad libitum* meal and self-reported intake, both groups were in positive compensation, with active demonstrating 35.6 ± 135.2% compensation and inactive demonstrating 3.0 ± 135.0% compensation (Jokisch *et al.* 2012). This study suggests that habitually active individuals are able to acutely (at the *ad libitum* meal) increase their energy intake while inactive decrease their energy intake. Additionally, the inactive group increased their energy intake throughout the remainder of the day, which suggests that inactive individuals might require a longer time frame to compensate for the exercise-induced energy deficit. These findings support previous evidence (King *et al.,* 1999; Long *et al.,* 2002; Martins *et al.,* 2013; Martins *et al.,* 2007b; Mayer *et al.* 1956; Van Walleghen *et al.,* 2007), suggesting a beneficial role for physical activity on the acute control of energy intake, however, more experimental studies are necessary to clarify and confirm these outcomes.

In summary, most studies have only examined the effects of an acute bout of high-intensity exercise on hunger and food intake in regular exercisers and from the few

studies examining inactive-to-moderately-active individuals there are mixed results. Furthermore, it is difficult for many inactive individuals to undertake and maintain high-intensity exercise programmes (Perri *et al.,* 2002); therefore, there is the need to examine the effects of an acute bout of exercise at an intensity suitable for this population (i.e. moderate-intensity) (Ekkekakis *et al.,* 2005). Other limitations in previous research include the use of *ad libitum* buffet-style meals (Harris & George, 2008; Jokisch *et al.,* 2012; Klausen *et al.,* 1999; Westerterp-Plantenga *et al.,* 1997) that are more appropriate for the assessment of macronutrient intake and food preference than energy intake (Blundell *et al.,* 2010). These types of meals present a variety of food cues that are likely to promote overconsumption (Norton *et al.,* 2006) through a delay in satiation and a sustained interest in different foods (Hetherington *et al.,* 2006). The estimation of energy expenditure through the American College of Sports Medicine metabolic equivalent equations (Jokisch *et al.,* 2012) is also a limitation as this is known to be less accurate in comparisons of energy expenditure among individuals (Glass & Dwyer, 2007) leading to a possible overestimation of energy expenditure (Cunha *et al.,* 2011). Moreover, some studies did not measure energy expenditure (Harris & George, 2008; Westerterp-Plantenga *et al.,* 1997) which limits the interpretation of their findings with regard to energy balance. Another limitation is the short observation period (within the same day) employed by four of these studies (Harris & George, 2008; Jokisch, *et al.,* 2012; Ueda *et al.,* 2009b; Westerterp-Plantenga *et al.,* 1997) which might not have been long enough to detect more delayed responses in the variables assessed. Therefore, there is a need to investigate the acute effects of exercise on hunger and food intake in inactive individuals over a longer period of time and with improved research designs. A better understanding of how exercise affects appetite and feeding behaviour during the

hours and days following exercise might provide insight into how to successfully manage possible compensatory responses.

2.6.1.2 Effects of acute exercise in women

Most studies investigating the effects of an acute bout of exercise on hunger and food intake in women reported that hunger does not change after an acute bout of exercise (Finlayson *et al.,* 2009; King *et al.,* 1996; Lluch *et al.,* 1998; Pomerleau *et al.,* 2004; Unick *et al.,* 2010) whilst in some studies hunger decreased (Reger *et al.,* 1984; Tsofliou *et al.,* 2003) and increased after exercise (Maraki *et al.,* 2005). Similarly to men, the subjective ratings of hunger in women did not translate to food intake immediately after exercise and most studies reported no significant effects of exercise on energy intake (Finlayson *et al.,* 2009; George & Morganstein 2003; Hagobian *et al.,* 2012; Lluch *et al.,* 1998; Lluch *et al.,* 2000; Maraki *et al.,* 2005; Larson-Meyer *et al.,* 2012; Tsofliou *et al.,* 2003; Unick *et al.,* 2010). Fewer studies have reported increases (Pomerleau *et al.,* 2004) and decreases (Kissilef *et al.,* 1990) on energy intake after exercise, however, when adjusting for the energy expended during exercise, several studies (Lluch *et al.,* 1998; Lluch *et al.,* 2000; Maraki *et al.,* 2005; Pomerleau *et al.,* 2004; Unick *et al.,* 2010) found a decrease in relative energy intake.

Similarly to most research examining the effects of an acute bout of exercise on food intake in active women, most studies with inactive participants reported no exercise-induced responses in energy intake (George & Morganstein, 2003; Maraki *et al.,* 2005; Reger *et al.,* 1984; Tsofliou *et al.,* 2003; Unick *et al.,* 2010) with only one study reporting a decrease in energy intake after exercise (Kissileff *et al.,* 1990). However, the definition of participants' inactivity in several of these studies was not stated (Reger *et*

al., 1984; Tsofliou *et al.,* 2003) or was too broad classifying participants exercising less than three times per week as non-regular exercisers (Kissileff *et al.,* 1990; Maraki *et al.,* 2005). Additionally, most of these studies (George & Morganstein, 2003; Kissileff *et al.,* 1990; Maraki *et al.,* 2005; Reger *et al.,* 1984) have examined premenopausal women without controlling variables such as the regularity of participants menstrual cycles, premenstrual or unusual menstrual symptoms, menstrual phase when testing and the use of hormonal contraceptive preparations which endangers the validity of their conclusions (Buffenstein *et al.,* 1995). This is reinforced in Vlitos and Davies (1996) review which defends that variation in cycle length, both between and within women, is one fundamental difficulty to overcome when considering the menstrual cycle. In another review of menstrual cycle and appetite research it was shown that from 30 studies comparing energy intake between cycle phases, 25 reported significantly higher energy intake at the luteal phase than follicular phase and that there is some evidence showing that women prone to premenstrual or unusual menstrual symptoms have greater fluctuations on energy intake and appetite (Dye & Blundell, 1997).

Previous research studies have also failed to report if participants were using oral contraceptives (OC) (George & Morganstein, 2003; Kissileff *et al.,* 1990; Maraki *et al.,* 2005; Reger *et al.,* 1984; Tsofliou *et al.,* 2003; Unick *et al.,* 2010). According to the United Nations the pill is the most common modem contraceptive method (including both reversible and non-reversible methods) in developed countries and the third most common in developing countries being used worldwide by 9% of women of reproductive age who are married or in a union (United Nations Department of Economic and Social Affairs, 2009). In Great Britain, 25% of women aged 16-49 years use OC and this percentage increases in particular age groups, 54% (aged 20-24), 41%

(aged 25-29) and 46% (aged 30-34) (Office for National Statistics, 2009). To the author's knowledge only one study (Hagobian *et al.*, 2012) examining the effects of exercise on appetite and energy intake reported that women were taking OCs.

While some studies have found that OC use increases food intake (Eck *et al.*, 1997; Naessen *et al.*, 2007) others have shown that non-oral contraceptive users and OC users describe similar patterns of food cravings and amount eaten across menstrual or OC cycle phases (Bancroft & Rennie, 1993; McVay *et al.*, 2011; Tucci *et al.*, 2010). Nevertheless, it is important to acknowledge that the study by Eck and colleagues (1997) used only triphasic oral contraceptives (contain three separate dosages of exogenous hormones, increasing throughout the 28 day period) and did not perform comparisons between non-OC users and OC users across cycle phases while the study by Naessen and colleagues (2007) compared energy intake before and after participants started using OC which taking into consideration that the body takes time to adapt after the beginning of OC, results might have been influenced by this initial period of adaptation. Moreover, an effect of OC on body mass is a common misconception among adolescents and even physicians (Hamani *et al.*, 2007) and recent Cochrane reviews showed that there is currently no evidence to support an effect of oral contraceptives on body mass (Gallo *et al.*, 2011) or any significant clinical advantage of biphasic and triphasic regimens over monophasics regimens in terms of safety or efficacy (Burkman *et al.*, 2011; Van Vliet *et al.*, 2011). Currently there is only some evidence of a depot-medroxyprogesterone acetate (DMPA) effect on body mass (Beksinska *et al.*, 2010; Berenson & Rahman, 2009) and therefore the exclusion of participants on DMPA is advisable in appetite research.

To the author's knowledge, only four studies (Hagobian *et al.,* 2012, Larson-Meyer *et al.,* 2012; Pomerleau *et al.,* 2004; Unick *et al.,* 2010) have attempted to control these variables, yet none of these studies controlled for premenstrual or unusual menstrual symptoms, three studies (Larson-Meyer *et al.,* 2012; Pomerleau *et al.,* 2004; Unick *et al.,* 2010) did not report if participants were taking oral contraceptives and the only study examining inactive participants (Unick *et al.,* 2010) did not control participants menstrual phases as testing occurred between days 7 and 21 of the menstrual cycle which when considering a 28-day menstrual cycle would mean that participants were tested during the mid- to late- follicular phase (days 7-13), ovulation (day 14) and early- to mid- luteal phase (days 15-21) (Jonge, 2003). Additionally, other limitations apply to these studies such as the use of *ad libitum* buffet-style meals (George & Morganstein, 2003; Reger *et al.,* 1984, Tsofliou *et al.,* 2003; Unick *et al.,* 2010), the estimation of energy expenditure using heart rate equations (George & Morganstein, 2003; Maraki *et al.,* 2005) and the lack of measurement of energy expenditure (Tsofliou *et al.,* 2003), which, as previously discussed, limits the interpretation of findings.

In summary, most studies have examined premenopausal women without controlling important factors such as the regularity of participants' menstrual cycles, premenstrual or unusual menstrual symptoms, the participants' menstrual phase when testing was completed and the use of hormonal contraceptive preparations. Moreover, interpretation of previous studies is often limited by participants' physical activity not being clearly defined, and by the methods used to assess energy intake and expenditure. Therefore, there is a need to better understand the effects of moderate-intensity exercise on hunger and food intake in active and inactive premenopausal women with better controlled studies. Moreover, the high prevalence of women using oral contraceptives demands a

better understanding of the effects of exercise on appetite and energy intake in women taking OCs.

2.6.1.3 *Effects of chronic exercise in men*

A single bout of exercise promotes different physiological changes from continued daily exercise sessions (Powell *et al.*, 2011) and chronic exercise has been reported to increase the sensitivity of appetite control (Martins *et al.*, 2013; Martins *et al.*, 2007b), therefore it is logical to assume that differences may arise between the individual and cumulative effects of exercise on appetite control. Additionally, a comprehensive understanding of the impact that exercise has on appetite control and energy balance can only be possible by researching the effects of acute and chronic exercise over various periods of time and with measurement of all energy balance components (Blundell *et al.*, 2012).

Although sex differences may exist in the control of appetite, men and women are frequently reported together when expressing the results from exercise interventions (Bryant *et al.*, 2012; Caudwell *et al.*, 2009; King *et al.*, 2008; King *et al.*, 2009; Koulouri *et al.*, 2006; Martins *et al.*, 2010; Martins *et al.*, 2013). Additionally, from the three studies (Caudwell *et al.*, 2009; King *et al.*, 2008; King *et al.*, 2009) that divided participants into two groups according to the actual to predicted change in body mass (i.e. classifying participants as responders if body mass changes were equal to or greater than the change expected due to the exercise-induced increase in energy expenditure and non-responders if body mass changes were less than the expected change), two did not report the number of participants in these final groups (Caudwell *et al.*, 2009) or the ratio of men and women (King *et al.*, 2008). The only study reporting the above

information (responders: 9 men and 23 women; non-responders: 10 men and 16 women) did not report the values for energy intake at 0 and 12 weeks for each group but rather that "*there was an increase in the energy intake at probe days of the non-responders (+164 kcal/d), but no change (or even a decline) in the energy intake of the responders (-125.9 kcal/d)*" (King *et al.,* 2009, p.924). Despite providing support that increased energy intake may be a major source of energy compensation in those individuals that did not lose the predicted amount of body mass (non-responders) (Caudwell *et al.,* 2009; King *et al.,* 2008; King *et al.,* 2009) they do not allow inferences about the effects of chronic exercise on appetite solely on men or women and therefore they will not be discussed in these sections.

Only a few studies have investigated the effects of chronic exercise on hunger in men (Caudwell *et al.,* 2013b; Guelfi *et al.,* 2012; Stubbs *et al.,* 2002a; Stubbs et al., 2004b; Whybrow *et al.,* 2008). When cycling exercise sessions were repeated over a period of 7 days hunger (assessed hourly during waking hours) did not change (Stubbs *et al.,* 2002a) or was reported to increase (Stubbs *et al.,* 2004b) compared with the resting condition in lean men. In a subsequent study, where six sedentary to moderately active lean men were studied during three 16 day protocols, corresponding to no exercise, two daily 40 min cycling sessions (to expend 28.6 kJ/kg body mass) and three daily 40 min cycling sessions (to expend 57.1 kJ/kg body mass), there were no differences in hunger between any of the conditions. These findings, were further supported by two studies examining the effects of 12 week exercise interventions in overweight and obese men, which reported no differences between area under the curve (AUC) hunger values at baseline and after week 12 (Caudwell *et al.,* 2013b; Guelfi *et al.,* 2012). Therefore, most evidence suggests that chronic exercise does not have an effect on hunger in men,

however, the length of these interventions ranged from 7 days to 12 weeks so it is not possible to ascertain if this lack of effect would persist with exercise interventions of longer duration.

In contrast with the limited research on hunger, several studies have examined the effects of chronic exercise on energy intake in men (Broeder *et al.,* 1992; Caudwell *et al.,* 2013a; Caudwell *et al.,* 2013b; Donnelly *et al.,* 2003; Drenowatz *et al.,* 2012; Frey-Hewitt *et al.,* 1990; Leon *et al.,* 1979; Martins *et al.,* 2007b; Staten, 1991; Stubbs *et al.,* 2002a; Stubbs *et al.,* 2004b; Van Etten *et al.,* 1997; Westerterp *et al.,* 1992; Whybrow *et al.,* 2008). Staten (1991) examined 10 men who ran 60 min a day on a treadmill at approximately 69% of V02max for 5 consecutive days. Energy intake increased by 875 kJ/d but this was still insufficient to fully compensate for the exercise-induced increase in energy expenditure (2495 kJ/d). This was further supported by Stubbs *et al.* (2004b) study where 8 lean men exercising for 120 min/d at mean relative intensity of 65% of V 02max partially compensated for the exercise-induced energy expenditure (4900 kJ/d) by increasing energy intake (+1200 kJ/d). Conversely, 7 days of moderate (80 min at a 44% of V02max per day) and high (120 min at 50% of V02max per day) levels of exercise did not lead to any compensation of energy intake in 6 lean men (Stubbs *et al.,* 2002a). Despite the lack of effect of exercise intensity on energy intake observed in the acute studies, it is possible that the same does not apply to chronic exercise and therefore the difference between these studies are possibly due to the differences in their relative intensity of exercise. A study comparing the dietary intake in 15 endurance-trained men during a week of high-volume (>14 h of training per week) and a week of low-volume training (<7 h of training per week), also reported no adjustment in energy intake in response to the different training periods (Drenowatz *et al.* 2012). However, in this study energy intake was assessed with an online food-frequency questionnaire and

estimated to be underreported by 45% and 39% of total daily energy expenditure during the high- and low-volume week, respectively.

With an increase in the duration of exercise interventions to 14 days, the degree of compensation seems to become more apparent despite high individual variability. In Whybrow *et al.* (2008) study six lean men exhibited a partial degree of energy intake compensation (two daily sessions: +1.0 MJ/d; three daily sessions: +1.4 MJ/d) to the energy expended in the exercise interventions (two daily sessions: -2.3 MJ/d; three daily sessions: -4.4 MJ/d). This suggests that exercise-induced energy expenditure acts as a driver of food intake, but that over these time periods (< 14 days), the increase in energy intake remains insufficient to fully compensate for the expended energy. Additionally, the increase in energy intake was associated with an increase in fluid intake, which according to the authors, suggests that the observed partial energy intake compensation may be due to the priority given to increasing fluid intake in the short to medium-term (Whybrow *et al.*, 2008).

These findings, are however not replicated in interventions of longer duration as a 6-week moderate exercise programme (four times per week, 65-75% maximal heart rate) failed to elicit any changes at a buffet lunch or in daily energy intake (assessed by 3-day food diaries) of 11 healthy previously sedentary men (Martins *et al.*, 2007b). Similarly, three other studies examining 12 weeks of supervised exercise in active (Broeder *et al.*, 1992) and overweight and obese men (Caudwell *et al.*, 2013a; Caudwell *et al.*, 2013b) reported no changes between pre- and post- intervention energy intake. Additionally, the decreases in participants' body mass observed in all of these studies also support this lack of effect of chronic exercise on energy intake.

Leon *et al.* (1979) examined the effects of 16 weeks of vigorous walking (90min, 5 days/week, on a treadmill at up to 5.2 km/h on a 10% grade) in 6 obese men. The mean daily energy intake (assessed by 3-day food diaries) was reported to increase from week 0 (9573 kJ/d) to week 4 (10812 kJ/d) and decrease progressively from week 4 to 16 of the exercise programme (week 8: 10289 kJ/d; week 12: 9874 kJ/d; week 16: 8991 kJ/d) (Leon *et al*., 1979). This study agrees with previous findings that have observed an initial partial energy intake compensation (Staten, 1991; Stubbs *et al.,* 2004b; Whybrow *et al.,* 2008) and, given how daily energy intake at baseline was closest to the energy intake at the 12th week, may explain why studies examining 12 weeks exercise interventions (Broeder *et al.,* 1992; Caudwell *et al.,* 2013a; Caudwell *et al.,* 2013b) did not observe any changes in daily energy intake. This finding does not explain the lack of change in Martins *et al.* (2007b) 6 weeks intervention study, however this could be explained by the differences in participants' characteristics (e.g. body fat) or intensity of exercise between these studies. Additionally, mean changes in energy intake in Leon *et al.* (1979) study are not able to fully account for changes in participants' body mass and therefore either participants underreported their energy intake or decreased their physical activity outside the exercise sessions. Unfortunately, it is not possible to ascertain if the latter could explain these findings as this study did not measure energy expenditure.

In contrast, exercise studies carried out over longer periods of time (> 4 months), in untrained non-obese (Frey-Hewitt *et al.,* 1990; Van Etten *et al.*, 1997; Westerterp *et al.,* 1992) and obese men (Donnelly *et al.,* 2003) reported no differences between pre- and post-intervention energy intake with only one study reporting a tendency for a decrease

in energy intake from month 5 to month 10 of the exercise intervention (Westerterp *et al*., 1992). These findings suggest that chronic exercise over long-periods of time either inhibits or does not affect energy intake. However, the latter cannot be confirmed as the long periods of time between measurements (e.g. 1 year between measurements in Frey-Hewitt *et al.,* 1990) makes adjustments in energy intake during this period not possible to detect and therefore impossible to exclude.

Collectively previous findings suggest that exercise interventions of up to 12 weeks do not have an effect on hunger in men. Most studies examining the effect of chronic exercise on energy intake support that, in the short-term, men can increase energy intake in response to chronic exercise but this is not sufficient to fully compensate for the exercise-induced energy expenditure. Comparisons of pre- and post-intervention energy intake in long-term studies suggest that energy intake does not track energy expenditure eliciting decreases in body mass, however, exclusion of possible changes during the intervention is not possible due to the long periods of time between measurements. Additionally, several studies did not provide clear definitions of participants' activity status (Broeder *et al.,* 1992; Caudwell et al., 2013b; Staten, 1991; Stubbs *et al.,* 2002a; Stubbs *et al.,* 2004b; Van Etten *et al.,* 1997; Whybrow *et al.,* 2008), did not supervise exercise sessions (Martins *et al.,* 2007b), did not measure energy expenditure (Leon *et al.,* 1979) and assessed energy intake through a food-frequency questionnaire (Drenowatz *et al.* 2012) which also limits interpretation of findings. It is also important to note that some of these studies (Broeder *et al.,* 1992; Leon *et al.,* 1979) were not specifically designed to determine an effect of chronic exercise on energy intake. Therefore, better controlled studies are needed to better understand the effects of exercise interventions on hunger and food intake in men.

2.6.1.4 Effects of chronic exercise in women

Similarly to men, only a few studies have investigated the effects of chronic exercise on hunger in women (Caudwell *et al.,* 2013b; Martins *et al.,* 2007b; Stubbs *et al.,* 2002b; Ueda *et al.,* 2013; Whybrow *et al.,* 2008). In a study comparing 7 days of a sedentary routine (i.e. no exercise), to moderate (two daily 40 min cycling sessions: 21.4 kJ/kg/d) and intensive exercise interventions (three daily 40 min cycling sessions: 42.8 kJ/kg/d), there were no changes in the subjective ratings of hunger (assessed hourly during waking hours) (Stubbs *et al.,* 2002b). However, when participants were asked to retrospectively assess their hunger with an end-of-day questionnaire a small increase was reported during the exercise interventions compared to the resting week (Stubbs *et al.,* 2002b). When exercise interventions were completed over a period of 14 days (Whybrow *et al.,* 2008), 6 weeks (Martins *et al.,* 2007b) and 12 weeks (Caudwell *et al.,* 2013b; Ueda *et al.,* 2013) no changes in hunger ratings were observed suggesting that, as in men, chronic exercise does not seem to have an effect on hunger in women during the short- (i.e. few days) and medium-term (i.e. few weeks).

Numerous studies have examined the effects of chronic exercise on energy intake in women (Caudwell *et al.,* 2013a; Caudwell *et al.,* 2013b; Church *et al.,* 2009; Donnelly *et al.,* 2000; Donnelly *et al.,* 2003; Foster-Schubert *et al.,* 2005; Irwin *et al.,* 2003; Keim *et al.,* 1996; Keim *et al.,* 1990; Manthou *et al.,* 2010; Martins *et al.,* 2007b; Snyder *et al.,* 1997; Staten, 1991; Stubbs *et al.,* 2002b; Suzuki *et al.,* 1998; Ueda *et al.,* 2013; Westerterp *et al.,* 1992; Whybrow *et al.,* 2008; Woo *et al.,* 1982a; Woo *et al.,* 1982b; Woo & Pi-Sunyer, 1985). From the studies examining exercise interventions of less than 3 weeks, two studies reported that chronic exercise increased energy intake (Stubbs *et al.,* 2002b; Woo & Pi-Sunyer, 1985) while four studies did not observe changes in

food intake (Keim *et al.,* 1996; Keim *et al.,* 1990; Staten, 1991; Whybrow *et al.,* 2008; Woo *et al.,* 1982a). Yet in one of these studies (Whybrow *et al.,* 2008) there was a tendency for women to increase their mean daily energy intake with moderate (+0.3 MJ/d) and high levels of exercise-induced energy expenditure (+1.6 MJ/d). Results from these studies suggest that the effect of exercise on food intake in lean women differs from that in obese women. From the previous studies investigating lean women (Staten, 1991; Stubbs *et al.,* 2002b; Whybrow *et al.,* 2008; Woo & Pi-Sunyer, 1985), only one study reported no trends or significant changes in energy intake (Staten, 1991), a finding that could be attributable to dietary influences as in this study participants were given a liquid meal replacement formula, and had a limited choice of food items to consume from. Conversely, the research studies with obese participants (Keim *et al.,* 1996; Keim *et al.,* 1990; Woo *et al.,* 1982a) reported no effect of chronic exercise on energy intake. Of interest is that when comparing two studies conducted with the same protocol in a metabolic ward, obese women did not increase food intake to compensate for the increased physical activity (Woo *et al.,* 1982a) whereas lean women did (Woo & Pi-Sunyer, 1985). Results of these two studies together suggest that unlike lean women, obese women's food intake is subject to a stronger influence of factors other than physical activity such as food-related cues (i.e. palatability, variety, and availability of food) or other sensory and psychological factors (Pi-Sunyer & Woo, 1985).

Despite the differences observed between lean and obese women in studies of up to 3 weeks, exercise interventions of 6 to 12 weeks did not elicit any changes between pre- and post-intervention lunch or mean daily energy intakes in lean (Martins *et al.,* 2007b; Suzuki *et al.,* 1998) and overweight and obese women (Caudwell *et al.,* 2013a; Caudwell *et al.,* 2013b; Manthou *et al.,* 2010; Ueda *et al.,* 2013; Woo *et al.,* 1982b).

These results suggest two possibilities; the differences between obese and lean women are more marked in the initial phase of an exercise intervention (i.e. first few weeks) and disappear as the body adapts to the exercise stimulus, or the observed differences between lean and obese participants are not due to body mass status per se but rather other participants characteristics not present in these studies.

Comparisons between lean and obese participants are difficult to make as the duration of exercise interventions increases because most long-term exercise intervention studies (> 6 months) have examined only overweight and obese women (Church *et al.,* 2009; Donnelly *et al.,* 2000; Donnelly *et al.,* 2003; Foster-Schubert *et al.,* 2005; Irwin *et al.,* 2003; Snyder *et al.,* 1997). Nevertheless, one study (Westerterp *et al.,* 1992) observed that lean women training for a half marathon had a tendency to increase their food intake from week 20 to week 40. This is possibly explained by the adjustment in participants running time made at week 20 from 10-30 min/d to 20-60 min/d (Westerterp *et al.,* 1992). Conversely, most studies examining overweight and obese women (Donnelly *et al.,* 2000; Donnelly *et al.,* 2003; Foster-Schubert *et al.,* 2005; Irwin *et al.,* 2003; Snyder *et al.,* 1997) reported no differences in energy intake between baseline and post-intervention suggesting an uncoupling of energy intake and energy expenditure in long-term exercise interventions. Nevertheless, in two of these studies (Donnelly *et al.,* 2000; Donnelly *et al.,* 2003) the body composition results suggested the presence of some form of compensatory response that, as discussed in a later report, is more likely to be due to changes in energy intake (Donnelly & Smith, 2005). A difference between the actual and predicted decrease in body mass was also observed in a study that reported that energy intake decreased after 6 months of three exercise interventions with different energy expenditures (17, 34 or 50 kJ/kg/week). In this study

the exercise intervention with higher energy expenditure (50 kJ/kg/week) only produced about half of the predicted weight loss. However, the use of food frequency questionnaires and step counters by this study does not allow confirmation of whether the observed compensation is the result of an increase in energy intake or a decrease in physical activity energy expenditure.

In summary, current evidence suggests that short- (i.e. few days) and medium-term (i.e. few weeks) exercise interventions do not exert a marked effect on hunger in women. From the short-term studies examining energy intake, a difference between lean and obese women was observed suggesting that in obese women, energy intake is strongly influenced by sensory and psychological factors. However, this difference is not observed when the length of the exercise interventions is increased to 6 weeks or more. Most long-term interventions (> 6 months) have reported no exercise-induced changes in energy intake, however, body mass data do not always support this suggesting that some form of compensation is present. Additionally, several methodological limitations are present in these studies such as not controlling women's menstrual cycle for the measurement of energy intake (Caudwell et al., 2013a; Caudwell et al., 2013b; Donnelly *et al.*, 2003; Keim *et al.*, 1996; Keim *et al.,* 1990; Manthou *et al.,* 2010; Martins *et al.,* 2007b; Stubbs *et al.,* 2002b; Suzuki *et al.,* 1998; Westerterp *et al.,* 1992; Whybrow *et al.,* 2008; Woo *et al.,* 1982a; Woo *et al.,* 1982b; Woo & Pi-Sunyer, 1985), not providing a clear definition of participants' activity status (Caudwell et al., 2013b; Keim *et al.,* 1996; Keim *et al.,* 1990; Snyder *et al.,* 1997; Staten, 1991; Stubbs *et al.,* 2002b; Whybrow *et al.,* 2008), not supervising exercise sessions (Martins *et al.,* 2007b; Snyder *et al.,* 1997), not measuring energy expenditure (Irwin *et al.,* 2003; Suzuki *et al.,* 1998; Ueda *et al.,* 2013) and assessing energy intake through a food-frequency

questionnaires (Church *et al.,* 2009; Foster-Schubert *et al.,* 2005; Irwin *et al.,* 2003). It is also important to note that several of these studies (Church *et al.,* 2009; Donnelly *et al.,* 2000; Foster-Schubert *et al.,* 2005; Irwin *et al.,* 2003; Jakicic *et al.,* 2003; Snyder *et al.,* 1997) were not specifically designed to determine an effect of chronic exercise on energy intake. Therefore, more research with improved measurement methods is needed to better understand the effects of exercise interventions on hunger and food intake in women.

2.6.2 Exercise and physical activity energy expenditure

Physical activity energy expenditure has been shown to contribute to energy balance and the control of body mass through its two components (i.e. exercise-induced energy expenditure and non-exercise energy expenditure) (King *et al.,* 2007). Therefore, the importance of energy expenditure in the energy balance equation demands that the focus on exercise-induced changes in energy intake does not overshadow the possible contribution of physical activity energy expenditure.

The possibility of an exercise-induced compensation in physical activity was first described by Rowland's (1998) "activity-stat" mechanism that proposed a central control of physical activity according to a set point of energy expenditure. An increase in physical activity would be later compensated for by a reduction in activity to preserve an individual's set point (Rowland, 1998). However, as exercise determines exercise-induced energy expenditure it can be assumed that, with the exception of failing to comply with the exercise intervention, exercise-induced compensatory changes are likely to occur in the non-exercise energy expenditure component. According to Levine (2004, p.E675), non-exercise energy expenditure or, as it is also known, non-exercise

activity thermogenesis (NEAT) is defined as "*the energy expendedfor everything that is not sleeping, eating, or sports-like exercise*" and similarly to eating behaviour, is believed to be biologically regulated and influenced or perhaps overridden by environmental cues (Levine & Kotz, 2005). Additionally, previous studies have demonstrated that changes in non-exercise energy expenditure mediate resistance to increases in body mass with overfeeding in sedentary adults (Levine *et al.,* 1999) and that low grade activities are sufficiently great to contribute to energy balance (Levine *et al.,* 2000). Therefore, it is possible that non-exercise energy expenditure contributes substantially to the inter- and intra-individual variability in energy expenditure (Levine, 2003) which might explain some differences in the effectiveness of exercise interventions.

2.6.2.1 Effects of acute and chronic exercise

To the author's knowledge only one study has examined the effects of a single bout of exercise on non-exercise physical activity in adults (Alahmadi *et al.,* 2011). Sixteen overweight and obese men performed a single bout of moderate- (60 minutes on a motorized treadmill at 6 km/h) and high-intensity walking (60 minutes of interval walking at 6 km/h; 10% grade for five minutes followed by five minutes at 0% grade) on two separate occasions. Non-exercise physical activity was assessed by accelerometers on the three days before each experimental day, on the exercise days and for three days after the experimental days (Alahmadi *et al.,* 2011). This study reported that a single bout of moderate- and high-intensity walking did not alter non-exercise physical activity on the exercise day or on the first 2 days, however on the third day after the exercise, an increase was observed with the high- but not with the moderate-intensity exercise session (Alahmadi *et al.,* 2011). These results suggest that an acute

bout of moderate- and high-intensity walking does not elicit a compensatory reduction in non-exercise physical activity in obese men and that high-intensity exercise might actually increase it. However, whilst this study compared two exercise interventions there was no control condition.

In contrast with the effects of a single exercise session, several research studies have examined the effects of chronic exercise on physical activity energy expenditure (Church *et al.,* 2009; Colley *et al.,* 2010; Goran & Poehlman, 1992; Hollowell *et al.,* 2009; Hunter *et al.,* 2000; Keytel *et al.,* 2001; Manthou *et al.,* 2010; McLaughlin *et al.,* 2006; Meijer *et al.,* 1991; Meijer *et al.,* 1999; Meijer *et al.,* 2000; Morio *et al.,* 1998; Racette *et al.,* 1995; Rosenkilde *et al.,* 2012; Stubbs *et al.,* 2002a; Stubbs *et al.,* 2002b; Stubbs *et al.,* 2004b; Turner *et al.,* 2010; Van Etten *et al.,* 1997; Wang & Nicklas, 2011). In a series of three studies with similar protocols (Stubbs *et al.,* 2002a; Stubbs *et al.,* 2002b; Stubbs *et al.,* 2004b) where participants exercised for 7 days, two studies reported that total daily energy expenditure (estimated from heart-rate) tended to decrease over time (7 days) with increasing exercise levels (Stubbs *et al.,* 2002a; Stubbs *et al.,* 2004b) while one reported no changes (Stubbs *et al.,* 2002b). Possible sex differences could explain the contrasting findings between these studies, however, this is not supported by a recent study that reported no changes in non-exercise energy expenditure (estimated from heart-rate and physical activity diaries) of 16 lean men (n=8) and women (n=8) exercising over 8 days (McLaughlin *et al.,* 2006).

Despite allowing the observation of the initial adaptations of physical activity energy expenditure to chronic exercise, these studies are limited by the short period of time over which the intervention is conducted. Results from exercise interventions of longer

duration (≥ 2 months) (Colley *et al.*, 2010; Goran & Poehlman, 1992; Hollowell *et al.*, 2009; Hunter *et al.*, 2000; Keytel *et al.*, 2001; Manthou *et al.*, 2010; Meijer *et al.*, 1991; Meijer *et al.*, 1999; Meijer *et al.*, 2000; Morio *et al.*, 1998; Racette *et al.*, 1995; Rosenkilde *et al.*, 2012; Turner *et al.*, 2010; Van Etten *et al.*, 1997; Wang & Nicklas, 2011) have also provided contradictory findings. Several studies have shown chronic exercise-induced decreases in non-exercise energy expenditure (Colley *et al.*, 2010; Goran & Poehlman, 1992; Manthou *et al.*, 2010; Meijer *et al.*, 1999; Meijer *et al.*, 2000; Morio *et al.*, 1998; Wang & Nicklas, 2011), whereas others reported no changes (Church *et al.*, 2009; Hollowell *et al.*, 2009; Keytel *et al.*, 2001; Turner *et al.*, 2010; Van Etten *et al.*, 1997), an increase (Hunter *et al.*, 2000; Racette *et al.*, 1995) and mixed findings between groups (Manthou *et al.*, 2010; Meijer *et al.*, 1991; Rosenkilde *et al.*, 2012). It is difficult to explain these conflicting findings, but it is likely that differences in intensity and type of exercise, measurement methods, and participants' characteristics such as age are contributing factors. For instance, two recent studies in overweight men (Rosenkilde *et al.*, 2012) and overweight or obese women (Wang & Nicklas, 2011) have suggested that exercise-induced reductions in non-exercise energy expenditure are dose-dependent. Rosenkilde et al. (2012) reported reductions only in individuals performing a high dose of exercise (2510 kJ/d) but not the ones performing a moderate dose of exercise (1255 kJ/d) while Wang and Nicklas (2011) reported decreases in non-exercise induced energy expenditure on the days of the exercise sessions with vigorous-intensity (70-75% of $\dot{V}O_{2max}$) but not on the days of moderate-intensity (45-50% of $\dot{V}O_{2max}$). Nevertheless, participants in the latter study were also on a constant caloric restriction diet and therefore results may only be applicable for women already restricting their diet. Additionally, another recent study conducted over a longer period of time (8 months) in overweight and obese men and women did not observe this dose-

response effect when comparing low amount/moderate-intensity, low-amount/vigorous-intensity, or high-amount/vigorous-intensity aerobic exercise (Hollowell *et al*., 2009). Therefore, uncertainty remains about a possible exercise-dose effect on changes in non-exercise induced energy expenditure.

Another possible explanation for differences between studies is participants' age as elderly individuals seem more likely to exhibit exercise-induced compensatory changes in non-exercise energy expenditure. Most studies in this population reported decreases (Goran & Poehlman, 1992; Meijer *et al.,* 1999; Meijer *et al.,* 2000; Morio *et al.,* 1998) and only one study reported an increase in non-exercise energy expenditure (Hunter *et al.,* 2000). This difference may be explained by the Hunter *et al.* (2000) study involving only resistance exercise in contrast with the aerobic (Morio *et al.,* 1998) and combined aerobic and resistance programs (Goran & Poehlman, 1992; Meijer *et al.,* 1999; Meijer *et al.,* 2000) delivered in the other studies. Although age-related differences are not possible to ascertain because no studies have directly compared younger to older adults, it is possible that the prevalence of these compensatory changes in this population are due to exercise being a more challenging stimulus in older than young adults.

The measurement of energy expenditure and/or physical activity is difficult due to its multiple domains (e.g. leisure, occupational and transportation) and dimensions (frequency, duration, intensity and type). Therefore, it is possible that differences between studies are a result of varying methods. There is currently no method available to measure all physical activity dimensions, however, a combination of heart-rate monitors and accelerometers is able to evaluate frequency, intensity and duration of physical activity over a specific period of time and is acknowledged to be a good

predictor of energy expenditure (Butte *et al.,* 2012; Jorgenson *et al.,* 2009) whereas self-reported physical questionnaires and logs are not (Foley *et al.,* 2012; Prince *et al.*, 2008). Nevertheless, several of the above studies have assessed physical activity through physical activity diaries (McLaughlin *et al.,* 2006; Manthou *et al.,* 2010; Morio *et al.,* 1998; Racette *et al.,* 1995) and pedometers (Church *et al.,* 2009).

In summary, to the author's knowledge, only one study has examined the effects of a single bout of exercise on non-exercise energy expenditure and reported an increase on the third day after vigorous but not moderate walking in obese men. However, this study did not have a control condition and used an ambiguous definition of participant's previous activity status *("All participants were sedentary and participated in less than 1 h/week of physical activity*") (Alahmadi *et al.,* 2011, p.625). Findings from the studies examining the effects of exercise interventions on non-exercise energy expenditure are equivocal. Differences might be explained by participants characteristics (e.g. age), exercise characteristics (e.g. intensity), and different measurement methods used (e.g. accelerometers vs. physical activity diaries), however, confirmation of these relationships is still not possible. Therefore, there is a clear need to investigate the acute and chronic effects of exercise on physical activity energy expenditure in active and inactive men and women using improved measurement methods (e.g. multisensor devices).

2.7 Aims

- To ascertain if compensatory changes in hunger, energy intake and/or energy expenditure occur after an acute bout of moderate-intensity exercise in active and inactive:
  - Men (study one; chapter 4)
  - Women not using hormonal contraceptives (study two; chapter 5);
  - Women taking oral contraceptives (study three; chapter 5);
- To ascertain if compensatory changes in energy intake and/or energy expenditure occur after 12 weeks of moderate-intensity exercise training in inactive men (study four; chapter 6).

Chapter 3: General methods

This chapter describes the experimental procedures that have been used in the following chapters of this thesis. All research studies were undertaken with the approval of Sheffield Hallam University's Faculty of Health and Wellbeing Research Ethics Committee (Appendix 1).

3.1 Participants

Participants were recruited from within Sheffield Hallam University and the local area by flyers, posters, e-mails and word of mouth. Before participation volunteers were given an information sheet with the purpose of the study, a description of the experimental procedures and the potential risks and discomforts associated with these procedures (Appendix 2). During the preliminary visit, participants were given the opportunity to ask any questions about the study after which they signed an informed consent document (Appendix 3). At this time, participants were also asked to complete a health screen questionnaire (Appendix 4), a physical activity questionnaire (Godin & Sheppard, 1985 - Appendix 5), an eating behaviour questionnaire (Karlsson *et al.* 2000 - Appendix 6), the Shortened Premenstrual Assessment Form (SPAF; Allen, McBride & Pirie, 1991 - Appendix 7, only on studies 2 and 3) and a food declaration form (Appendix 8) to ensure participants were not allergic to any of the ingredients present in the meals provided during the study. Inclusion criteria for participant recruitment are presented separately in each chapter due to being specific to each research study.

3.2 Eating behaviour

Participants were asked to complete the Three-Factor Eating Questionnaire (TFEQ-R18) (Karlsson *et al.,* 2000 - Appendix 6) during the preliminary visit. The TFEQ-R18 was developed on the basis of psychometric analyses of the original 51-item TFEQ in a sample of 4377 Swedish, middle-aged, obese men and women (Karlsson *et al.,* 2000). Although it was constructed using data from obese adults, it has been shown to be valid in the general population in France (De Lauzon *et al.,* 2004) and in the United States (Cappelleri et al., 2009), and in female adolescents and adults in Finland (Angle *et al.,* 2009). The questionnaire is composed of 18 questions that measure 3 different aspects of eating behaviour: restrained eating (conscious restriction of food intake in order to control body mass; 6 items), uncontrolled eating (tendency to eat more than usual due to a loss of control over intake accompanied by subjective feelings of hunger; 9 items) and emotional eating (inability to resist emotional cues; 3 items). As recommended by Karlsson *et al.* (2000), responses to statements were done on a 1-4 Likert scale (1= definitely false, 4 = definitely true) with the values being initially summated into scale scores for cognitive restraint, uncontrolled eating, and emotional eating, and later converted to a 0-100 scale using the following formula:

(Raw score - lowest possible score) / (possible raw score range) x 100.

The ranges for the items were 6-24 for cognitive restraint, 9-36 for uncontrolled eating, and 3-12 for emotional eating with higher scores in the respective scales being indicative of greater cognitive restraint, uncontrolled, or emotional eating. As high levels of cognitive restraint have been associated with intent to diet and controlled eating (Lawson *et al.,* 1995) and reported to have strong associations with energy

underreporting (Maurer *et al.,* 2006) all studies presented in this thesis excluded highly restrained participants. However, it is important to consider that this scale was not designed to measure disordered eating behaviour, and does not designate a cut-off score that would indicate the threshold of clinical levels of restrained eating. Therefore, participants were deemed as highly restrained eaters when scoring more than 18 (raw score out of a total of 24) or 66% (percentage score). In the longitudinal study (chapter 6) these values were measured before and after the exercise intervention to ascertain if changes occurred over this period of time.

3.3 Anthropometry

Participant's anthropometric characteristics were determined at the preliminary visit according to International Society for the Advancement of Kinanthropometry guidelines (ISAK; Marfell-Jones *et al.,* 2006).

3.3.1 Stature

Stature was measured to the nearest 0.01 m using a wall-mounted stadiometer (Harpenden, Holtain Ltd, Crymmych, Wales). Participants stood barefoot with their heels together, back against a wooden plate and arms by their side. The head was erect and in the Frankfort horizontal plane i.e. a horizontal plane represented in profile by a line between the lowest point on the margin of the orbit and the highest point on the margin of the auditory meatus. The stadiometer moveable head plate was then lowered to the top of the head at which point the participant was asked to breathe in deeply. The measurement was recorded just before the participant exhaled.

3.3.2 Body mass

Body mass was measured using a balance beam scale (model 424; Weylux; Hallamshire Scales Ltd, Sheffield, UK) to the nearest 0.05 kg. All participants were measured wearing light clothing with all footwear, jewellery and accessories/items in pockets being removed prior to the measurement. Measurement was taken with participants standing still, both feet flat and hands by their side.

3.3.3 Body mass index

Body mass index (BMI) was calculated as body mass in kilograms divided by the square of height in meters.

3.3.4 Circumferences

Waist and hip circumferences were measured directly on the skin using a steel flexible tape (Rosscraft Innovations Inc., Surrey, Canada) while the participant stood with arms at the sides, feet together and the abdomen relaxed. Measurements were taken at the end of a normal expiration with minimal skin compression. Waist circumference was measured at the narrowest part of the torso, above the umbilicus and below the xiphoid process. Hip circumference was measured at the maximal circumference of the buttocks. Waist circumference was divided by hip circumference to determine the waist-to-hip ratio.

3.3.5 Skinfolds thicknesses

Measurements of subcutaneous fat were performed in the cross-sectional studies (chapters 4 and 5) to estimate total body fatness. Skinfold thickness was measured to the nearest 0.2 mm using a Harpenden skinfold calliper (Baty International, Burgess Hill, UK). The measurements were made at the following sites on the right hand side of the body with the participant standing upright in the anatomical position:

1) Tricep — Vertical fold, on the posterior midline of the upper arm, halfway between the acromion and the olecranon processes, with the arm held freely to the side of the body.

2) Bicep — Vertical fold, on the anterior aspect of the arm over the belly of the biceps muscle, 1cm above the level used to mark the tricep site.

3) Subscapular - Diagonal fold (45° angle), 1 to 2 cm below the inferior angle of the scapula.

4) Suprailiac — Diagonal fold, in line with the natural angle of the iliac crest taken in the anterior axillary line immediately superior to the iliac crest.

Each skinfold was lifted by the experimenter's left hand thumb and index finger. The skinfold caliper was placed 1 cm away from the thumb and finger, perpendicular to the skinfold. The measurement was taken after 1-2 seconds of calliper pressure while still pinching the skinfold. A rotation through measurement sites was used to avoid any unnecessary compression of the adipose tissue and subsequent underestimation of skinfold thickness. The mean of three measurements for each site was calculated and used to represent the skinfold thickness for that site. The sum of skinfolds was used to

calculate body density using the predictive equations of Dumin and Womersley (1974) which was then used to estimate body fat percentage using the Siri equation (Siri, 1956).

3.3.6 Body composition

Body fat percentage, fat mass (FM), skeletal muscle mass (SMM) and fat free mass (FFM) were obtained via a bioelectrical impedance body composition analyser InBody720 (Derwent Healthcare Ltd, Newcastle upon Tyne, UK). Body fat percentage was recorded to the nearest 0.1 %, and FM, SMM and FFM to the nearest 0.1 kg. This instmment uses eight tactile electrodes, two in contact with the palm and thumb of each hand and two with the anterior and posterior aspects of the sole of each foot. These four pair of electrodes allows five segmental impedances (right arm, left arm, right leg, left leg and trunk) to be measured for 1, 5, 50, 250, 500 and 1000 kHz, thereby combining upper-, lower-, and total-body bioelectrical impedance to estimate FFM from the summation of intracellular water, and extracellular water. Body fat mass was calculated by excluding FFM from total body mass and SMM is estimated from total body water based on the assumption that hydration of fat-free mass is 73.2%. Before measurements the skin and the electrodes were cleaned using the specific electrolyte tissue according to the manufacturer's instructions. Measurements were performed without shoes and socks with participants being instructed to slightly abduct their arms and remain still in the upright position. All measurements were performed with the participants being at least two hours fasted and without having engaged in any kind of exercise during that day.

3.4 Arterial blood pressure

Arterial blood pressure was measured during health screening by a calibrated oscillometric blood pressure monitor (Dash 2500, GE Healthcare, Finland) according to the practice guidelines of the European Society of Hypertension (O'Brien *et al.* 2005). In the cross-sectional studies (chapters 4 and 5) all measurements were taken with participants seated for at least five minutes, relaxed and not moving or speaking whereas in the longitudinal study (chapter 6) participants lied down for 10 minutes, relaxed and not moving or speaking. The arm being measured was supported at the level of the heart and was not constricted by tight clothing. Measurements were taken in duplicate and the mean of these values recorded.

3.5 Heart rate

Heart rate during exercise was determined using short range telemetry (Polar F4, Polar Electro, Kempele, Finland) (chapters 4, 5 and 6) whereas during rest it was determined using a calibrated oscillometric blood pressure monitor (Dash 2500, GE Healthcare, Finland) (chapter 6). During the exercise intervention, exercise intensity (i.e. target heart rate) was prescribed as percentage of heart rate reserve (HRR) which was calculated as follows:

Age-predicted maximum heart rate = 205.8 – 0.685 × (age) (Inbar *et al.*, 1994)

Heart rate reserve (HRR) = maximum heart rate – resting heart rate

Target heart rate = (HRR × percentage of training intensity) + resting heart rate

3.6 Ratings of perceived exertion

Ratings of perceived exertion (RPE) were used to obtain each participants perception of exercise intensity using the Borg Scale (Borg, 1973), with numbers ranging from 6 (no exertion at all) to 20 (maximal exertion).

3.7 Analysis of expired air

Expired air collections were made in the cross-sectional studies (chapters 4 and 5) using two methods. The open-circuit spirometry method was used for the determination of maximal oxygen consumption. The Douglas-bag method was used to estimate experimental days energy expenditure through indirect calorimetry.

3.7.1 Determination of maximal oxygen consumption

A MedGraphics CPX Ultima (Medical Graphics Ltd, Gloucester, UK) gas-analysis system was used for the determination of maximal oxygen consumption. This device uses a mouthpiece and generates breath-by-breath data. Data generated was used to calculate mean values for every 20 seconds and included oxygen consumption ($\dot{V}O_2$), carbon dioxide production, expired minute ventilation and respiratory exchange ratio (RER). Calibration of the Ultima was performed immediately before each test using a 3-liter syringe and calibration gases of known concentration of oxygen [O_2 (12% / 21%)] and carbon dioxide [CO_2 (5%)]. The maximal value for oxygen consumption was taken as the highest 20-second mean value obtained during the last stage of the exercise test. To ensure that a true cycling-specific maximal oxygen consumption had been attained, two or more of the following criteria were met: participant heart rate within 15

beats/min of age-predicted maximum heart rate (205.8-0.685(age)) (Inbar *et al.,* 1994), an increase in VO_2 of less than 100 ml-min'1 despite an increase in exercise intensity, and a RER greater than 1.15.

3.7.2 Indirect calorimetry

Expired air samples were collected using in 150 1 Douglas Bags (Harvard Apparatus, Edenbridge, Kent, UK) and analysed using an oxygen/carbon dioxide gas analyser (Dual Gas Analyser GIR250, Hitech Instruments, Luton, UK). This dual sensor analyser was calibrated before each analysis by zeroing both oxygen and carbon dioxide sensors with 100% nitrogen (MaxiCan, Spantech, Surrey, UK) and then spanning the oxygen and carbon dioxide sensors with 4% carbon dioxide and 16% oxygen (MaxiCan, Spantech, Surrey, UK). A dry gas meter (Harvard Apparatus, Edenbridge, Kent, UK) was used to determine expired air volumes which were corrected to standard temperature, pressure and dry gas. Indirect calorimetry was used to determine energy expenditure from the above values based on the equations described by Frayn (1983).

3.8 Exercise tests

All participants completed the preliminary exercise protocols on a cycle-ergometer (Model 874E, Monark, Sweden). In the cross-sectional studies (chapters 4 and 5) participants completed a submaximal followed by a maximal cycle-ergometer test to calculate the cycling exercise intensity-oxygen consumption relationship and estimate their maximum oxygen consumption. In the longitudinal study (chapter 6) participants'

maximum oxygen consumption was estimated using the Astrand-Rhyming submaximal cycle-ergometer test.

3.8.1 Submaximal cycle-ergometer test

Before each test participants were allowed time to warm-up. After completing this, participants started a submaximal-incremental cycle-ergometer test to determine the relationship between cycling intensity and oxygen consumption. This test was designed to exercise participants through a range of intensities from moderate to vigorous, but not maximum. The test consisted in a maximum of 16 min of continuous cycling divided into four, 4-min stages or until the participant reached a heart rate of 160 bpm. The pedalling rate was initially set at 60 rpm (revolutions per minute) but participants were allowed to choose a different rate if they felt uncomfortable or could not maintain this cadence. Initial resistance was adjusted to the individual activity level with inactive participants starting at 9.8 N [1.0 kilogram-force (kgf)] and active at 14.7 N (1.5 kgf) or 19.6 N (2.0 kgf). At the end of each 4 min stage there was an increase of the resistance by 4.9 N (0.5 kgf). Participants were required to do the entire test while maintaining a sitting position. A MedGraphics CPX Ultima (Medical Graphics Ltd, Gloucester, UK) gas-analysis system was used to determine oxygen consumption and carbon dioxide production. A heart rate monitor (Polar F4, Polar Electro, Kempele, Finland) was used to assess each participant's heart rate continuously for every 15 seconds during the last minute of each stage. In addition, ratings of perceived exertion (Borg, 1973) were assessed during the same time periods. At the end of the test, the cycling exercise intensity-oxygen consumption relationship was determined by plotting oxygen consumption and exercise intensity data from each stage.

3.8.2 Cycle-ergometer maximum oxygen uptake test

After 20–30 min recovery, participants began the cycle-ergometer maximum oxygen uptake test. The test involved cycling continuously through 3-min stages until volitional exhaustion occurred. The preset pedalling rate was the same as the one chosen for the submaximal test and initial resistance was set to elicit a heart rate of 160 bpm based on data from the submaximal cycle ergometer test. At the end of each 3-min stage there was an increase of the resistance by 4.9 N (0.5 kgf). Strong verbal encouragement was given to participants throughout the test which was terminated when the participant failed to maintain cycling cadence for 20 consecutive seconds or signalled as not being able to continue. The MedGraphics CPX Ultima (Medical Graphics Ltd, Gloucester, UK) gas analysis system was used for the determination of maximal oxygen consumption. A heart rate monitor (Polar F4, Polar Electro, Kempele, Finland) was used to assess each participant's heart rate continuously and record it every 15 seconds during the last minute of each stage. Ratings of perceived exertion (Borg, 1973) were also assessed during the same time periods.

Once the test was finished, the participants' maximum oxygen consumption value was considered together with the data obtained during the submaximal test to determine cycling intensity to be used during the exercise experimental days, i.e. exercise intensity necessary to elicit 50% of their maximum oxygen consumption.

3.8.3 Astrand-Rhyming submaximal cycle-ergometer test

The Astrand-Ryhming cycle ergometer test (Astrand & Rhyming, 1954) was used for the estimation of participant's maximum oxygen consumption in the longitudinal study

(chapter 6). The test consisted of 6 min of continuous cycling with the goal to obtain a HR between 125 and 170 bpm. The pedalling rate was initially set at 50 or 60 rpm depending on participant's ability to maintain it. Exercise intensity was adjusted to the individual as suggested by the test protocol. After completion of the test, the resistance of the cycle-ergometer was decreased and participants cooled down. The mean of the last two HR and the final exercise intensity was then used to estimate maximum oxygen consumption from the adjusted nomogram (Astrand, 1960).

The Astrand-Rhyming submaximal test has a reported standard error of estimates from directly measured maximal oxygen consumption of ±10% for well-trained individuals and ±15% for untrained individuals (after use of the age-correction factor) and a test-retest correlation of 0.96 (Macsween, 2001). These differences can be due to the test underlying assumptions, i.e. a linear relationship between HR and $\dot{V}O_2$, maximum HR at a given age is constant and mechanical efficiency (oxygen consumption at a given exercise intensity) is the same for everyone.

3.9 Environmental temperature, humidity and pressure

The environmental temperature and humidity were determined using a digital thermo-hygrometer (ETHG 880, IDT International, Hong Kong). Barometric pressure was obtained by using a Fortin mercurial barometer.

3.10 Dietary and physical activity standardisation

In the cross-sectional studies (chapter 4 and 5) participants were asked to refrain from consuming alcohol or caffeine and taking part in vigorous physical activity in the 24

hours before each experimental day. Additionally, participants recorded their food intake on the two days before the first experimental day. This allowed participants to keep their activity patterns consistent between conditions and replicate their food intake during the two days before the second experimental day.

3.11 Subjective ratings of hunger

Subjective ratings of hunger were assessed with 100 mm visual analogue scales (VAS - Appendix 9) during the cross-sectional studies (chapters 4 and 5) as these have been shown to be a valid and reliable method for measuring appetite (Flint *et al.,* 2000; Merril *et al.,* 2002). VAS were preceded by a question (e.g. how hungry do you feel?) and anchored on the left and right by two expressions indicating its direction (e.g. "not at all hungry", "very hungry") (Flint *et al.,* 2000). Participants placed a vertical mark through the line at the point which best matched their present feeling of hunger. The distance from the left anchor to the vertical mark was then used as the hunger score. Subjective ratings of happiness, tiredness and nausea were included as distractions from the main outcome. This was important because participants were blinded to the true purpose of the studies (effects of an acute bout of exercise on immediate and subsequent three-day energy intake and expenditure) and were informed that the purpose of the investigation was to assess how food and physical activity affected mood to minimise participant-expectancy effects.

3.12 Breakfast and *ad libitum* lunch meal

Nutritional information for the cross-sectional studies (chapters 4 and 5) meals is presented in table 3.1. Breakfast was standardised across conditions and quantities were

determined based on individual body mass with participants consuming approximately 23.61 kJ/kg of body mass. Breakfast consisted of a bowl of cereal (Corn Flakes, Kellogg's, UK) with fresh semi-skimmed milk (Sainsbury, UK) and a glass of UHT orange juice (Drink Fresh, DCB Foodservice, UK). The *ad libitum* lunch meal consisted of durum wheat semolina conchiglie pasta (Granaria, Favellato s.r.l, Italy) served with tomato and mascarpone cheese sauce (Fratelli Sacla, S.p.A., Asti, Italy), and the combined meal comprised 10.1% energy from protein, 67.2% carbohydrate and 22.7% fat, with an energy density of 7.4 kJ/g. Cooking and cooling times were standardised across conditions to ensure a consistent carbohydrate availability across conditions and the pasta and sauce meal was served on both experimental days at a temperature of 60-65°C.

Table 3.1 Nutritional composition of breakfast and *ad libitum* lunch meals

Meal	Protein (%)	CHO (%)	Fat (%)	Energy density (kJ/g)
Breakfast	13	77	10	3.5
12% Corn Flakes	8	90	2	15.6
46% Semi-skimmed Milk	28	41	31	2.1
42% UHT Orange Juice	7	93	0	1.7
***Ad libitum* Lunch**	10	67	23	7.4
75% Conchiglie Pasta	12	84	4	14.9
25% Tomato and Mascarpone Cheese Sauce	3	14	83	7.0

CHO = carbohydrates.

3.13 Laboratory energy intake

Participants ate their breakfast and *ad libitum* lunch alone in individual air-conditioned testing cubicles equipped with Sussex Ingestion Pattern Monitors (SIPM), which consists of a concealed digital balance (KMB-TM, Kern, Germany) connected to a PC computer. These were developed at the University of Sussex (Yeomans, 2000), based on a modification of the Universal Eating Monitor (Kissileff *et al*., 1980), and have been used extensively in studies of human appetite (Bertenshaw *et al.,* 2009; Yeomans *et al.,* 2009). The system was custom programmed by the experimenter to give participants instructions (e.g. "please place the cutlery on the provided side plate when not eating") and was able to covertly monitor food intake to a precision of O.lg. This required participants to interact with a touch screen system and therefore a training period was given to them before breakfast to ensure no doubts would arise at the lunch meal. At lunch participants were given a 400g plate of pasta and instructed to "eat as much or as little as you want". To ensure participants did not use the empty plate as an external cue to end their meal, the SIPM was programmed to prompt the participant to call the experimenter, using a call button, once at least 300 g was consumed to receive a refill. This process was repeated until the participants indicated that they had finished eating. A separate side plate was provided for the participant to place cutlery on when not eating with it (e.g. still chewing food) to ensure the weight of cutlery did not interfere with the weighing process. The crockery and cutlery were standardised across conditions. Additionally, all bowls of food were separately weighed in the kitchen with a digital balance (KMB-TM, Kern, Germany) before and after consumption and during refills to ensure that the values provided by the SIPM were correct.

3.14 Free-living energy and macronutrient intake

As previously mentioned, the difficulty of obtaining accurate estimates of energy intake in the free-living is a main limitation in this field. Each method has notable strengths and limitations to be considered, and the most appropriate method depends on several factors such as the objectives of the study, the available resources and the population of interest (Johnson, 2002). Self-reported food diaries are a flawed but adequate method to measure energy intake in the free-living (De Castro, 2010) and self-reported weighed diet records have been historically considered the 'gold standard' for examining free-living energy and macronutrient intake (Black *et al.,* 1991). Therefore, in the studies presented in this thesis participants were instructed to weigh and record all items of food and drink consumed both at home and outside the home in food diaries (Appendix 10). All participants received guidance on how to complete the dietary record and measure food portions and instructed to contact the experimenter if they were unsure or had any questions about how to record foods accurately. When weighing was not possible, participants were asked to estimate portion sizes using standard household measures. Immediately upon receipt, food diaries were reviewed in the presence of the participant to ensure completeness and legibility, with any missing or unclear items being corrected. Food diaries were analysed to estimate energy and macronutrient intake using the dietary analysis software NetWisp (version 3.0; Tinuviel Software, Warrington, UK) with unlisted foods being inputted according to their package nutritional label.

According to Black *et al.* (1991), 7-day food diaries offer the best compromise between accuracy, researcher workload and participant compliance, however, this type of diaries was only applied in the longitudinal study (chapter 6) with 4-day food diaries being

used in the cross-sectional studies (chapters 4 and 5) due to their short-term duration. It is important to note that dietary records are prone to bias, usually towards under-reporting of energy and misreporting of macronutrients due to different forms of behaviour such as food being deliberately not reported (intentional under-reporting), food consumption being altered during the period of recording due to the burden of self-report and food being eaten but genuinely forgotten (unintentional/unknowing under-reporting) (Macdiarmid & Blundell, 1998). Therefore, the ratio of the reported daily food energy intake (EI) to the BMR was calculated for each participant (EI:BMR) to assess possible underreporting. The cut-off used for identifying unrepresentative intake was EI:BMR of 1.1 (Black *et al.,* 1991; Goldberg *et al.,* 1991).

3.15 Free-living energy expenditure

Free-living energy expenditure was estimated using an Actiheart (Cambridge Neurotechnology, Cambridge, UK), which is a single-piece light-weight (-10 g) water-proof device combining a heart rate monitor with an accelerometer (Brage *et al.,* 2005). The Actiheart uses a branched equation modelling meaning that the contribution of heart rate and accelerometer data towards the estimation of energy expenditure above rest is weighed differently depending on *a priori* established accelerometry-physical activity intensity and heart rate-physical activity intensity relationships (Brage *et al.,* 2004) (Figure 3.1). This device has been previously examined in the laboratory setting where it provided accurate estimates of low-to-moderate activities (Thompson *et al.* 2006) and was considered to be a reliable and valid tool for the measurement of movement and heart rate in adults (Brage *et al.,* 2005). Recently the Actiheart has been

considered valid for measuring and categorising intensities of physical activity in free-living individuals (Barreira *et al.*, 2009; Crouter *et al.*, 2008).

The Actiheart was attached to the participant's chest using two electrocardiogram (ECG) electrodes (E4 T815 Telectrode, Surrey, UK), a medial electrode placed at the level below the apex of the sternum and a lateral electrode placed on the same horizontal level as lateral as possible (lower position shown in figure 3.2). This positioning of the monitor at the level below the apex of the sternum was chosen because it is associated with cleaner heart rate data, particularly in men (Brage *et al.*, 2006) and was preferred by a pilot group due to the device being less visible. Participants were told to wear the monitor at all times, when awake or asleep and in study four participants were also asked to record the times (if any) where they did not wear the Actiheart in an activity log (Appendix 11). The epoch (i.e. interval of time between recordings) was set for 15 seconds in the cross-sectional studies (chapter 4 and 5) and 1 minute in the longitudinal study (chapter 6).

At the end of the free-living period, participants returned the Actihearts and the data were downloaded using a docking station and analysed using its commercial software. Heart rate and accelerometer data were converted to energy expenditure using Brage *et al.* (2007) revised group calibration branched equation. Total daily energy expenditure was calculated as the sum of physical activity energy expenditure (PAEE), diet-induced thermogenesis, and resting energy expenditure. Resting energy expenditure was estimated from the Schofield equations (Schofield, 1985) while diet-induced thermogenesis was assumed to equal 10% of total energy expenditure.

Figure 3.1 Representation of the branched model equation combining accelerometry and heart rate data adapted from the Actiheart user manual (2010). Pi - P_4 are weighting factors. X is used as the threshold to discriminate between activity and “no activity”. Y and Z are used to apply heart rate thresholds in the presence and absence of activity, respectively, and Y is used to discriminate between walking and running. At running speeds heart rate is a very reliable measure of energy expenditure whereas activity as measured by vertical acceleration is less reliable since during running the latter does not increase linearly with speed. This is reflected by the weighting in equation 1 where PI is high. At the other end of the spectrum (equation 4), heart rate is a poor measure of intensity whereas movement registration is more reliable and this is reflected by a relatively low weighting of the heart rate-energy expenditure relationship, that is, P_4 is low. Z is used to discriminate between raised heart rate due to some true activity in the presence of “no activity” (as set by X) and raised heart rate due to other factors. In equations 2 and 3 accelerometer movement and heart rate are equally weighted. The heart rate-physical activity intensity relationship and the accelerometry-physical activity intensity relationships used are given by the chosen calibration regression.

Figure 3.2 Two possible positions of the Actiheart. The upper position is at the level of the third intercostal space and the lower position is just below the apex of the sternum.

3.16 Statistical analyses

Statistical Package for the Social Sciences program for windows (SPSS 19.0, Chicago, IL) was used for all analyses. Data were checked for normal distribution using histograms and Shapiro-Wilk tests. Homogeneity of variance and sphericity were checked using Levene's and Mauchley's test, respectively. Results are reported as mean and standard deviation (mean ± SD) unless specified otherwise. Statistical significance was accepted at the 5% level.

3.16.1 Cross-sectional studies statistical analyses (Chapters 4 and 5)

Area under the curve (AUC) values for hunger were calculated using the trapezoidal rule. Net exercise-induced energy expenditure was calculated as (energy expenditure during the 60 min cycling period - energy expended during equivalent control period). Relative energy intake was calculated as (lunch energy intake condition) - (energy expended during the 60 min period of condition). Differences between groups for baseline characteristics, relative intensity of exercise (% of VC^max), ratings of perceived exertion (RPE) during exercise and net energy expenditure of the cycling period were assessed by independent Student's t-tests. The percentages of energy compensation identified the magnitude of possible compensations, therefore only group comparisons were made using a one-way ANOVA with the Welch test (when homogeneity of variance was violated). Two-way mixed-model ANOVAs (Group x Condition) compared the experimental day's lunch energy intake, energy expenditure, heart rate and respiratory exchange ratio (RER). Three-way mixed-model ANOVAs (Group x Condition x Time) compared mean daily energy and macronutrient intakes. In these analyses energy intake on the experimental day was calculated by summing participants' energy intake throughout the day (breakfast + *ad libitum* lunch + remainder of experimental day). However, the same formula could not be applied to macronutrient intake because the macronutrient values for breakfast and lunch of the experimental day were fixed. Therefore, macronutrient intake for the experimental day is limited to the free-living period of that day (i.e. remainder of the experimental day). Subjective hunger ratings and daily energy expenditure were also analysed with three-way mixed model ANOVAs (Group x Condition x Time). Where appropriate, post hoc tests were performed using Bonferroni adjustments. Cohen's *d* (standardised mean difference) effect sizes were calculated by dividing the difference between means by the pooled

standard deviation thus reflecting differences expressed in standard deviation units. According to Cohen's (1988) guidelines, effect sizes may be conservatively interpreted as small (0.2), medium (0.5), and large (0.8) effects. In addition, 95% confidence intervals were determined for energy intake, macronutrient intake, energy expenditure and percentage of energy compensation.

3.16.2 Longitudinal study statistical analyses (Chapter 6)

Paired t-tests compared estimated exercise energy expenditure, body composition, resting heart rate, blood pressure, estimated maximum oxygen consumption, metabolic profile (total cholesterol, HDL, non-HDL, triglycerides, LDL, and fasting glucose), cognitive restraint, unrestrained eating, emotional eating and food cravings before and after the exercise intervention. Two-way repeated measures ANOVA (Intervention x Time) compared energy intake, macronutrient intake and energy expenditure before and after the exercise intervention (Intervention effect) over the 7 days (Time effect). Cohen's *d* (standardised mean difference) effect sizes were calculated by dividing the difference between means by the pooled standard deviation thus reflecting differences expressed in standard deviation units. According to Cohen's (1988) guidelines, effect sizes may be conservatively interpreted as small (0.2), medium (0.5), and large (0.8) effects.

3.17 Reliability studies

One essential principle of the scientific method is that experiments must produce reproducible data to be meaningfully interpreted, therefore, reliability is an essential source of validity evidence for scientific data (Downing, 2004). Irrespectively of the

chosen equipment or operator doing the measurement, there are always internal and external factors that lead to some measurement error (Mullineaux *et al.*, 1999). Hence, to ensure confidence in the results, there is a need to quantify and minimise this measurement error whose main components are systematic bias (e.g. learning effect) and random error (biological or mechanical variation) (Ortega *et al.* 2008). In this regard, two reliability studies (one for male participants and one for female participants) were undertaken to evaluate the test-retest reproducibility of measurements obtained in this thesis. Twenty participants (10 men and 10 women) attended two assessments, one week apart, at approximately the same time of day (there was less than one hour difference in time of day between assessments) to minimise circadian or similarly induced variations. Participants were asked to refrain from consuming alcohol or caffeine and taking part in vigorous physical activity in the 24 hours prior to both tests. As no statistical test is singularly ideal as a measure of reliability, intraclass correlation coefficient (ICC - calculated using a 2-way random effects absolute agreement model) (Weir, 2005), technical error of measurement (TEM), coefficient of reliability (R) (Goto & Mascie-Taylor, 2007) and typical error (TE - also known as standard error of the measurement) (Atkinson & Nevill, 1998; Hopkins, 2000) were calculated with the results of the two visits in men (Table 3.2) and women (Table 3.3). Definitions and interpretations of these statistical tests are presented below:

- The intraclass correlation coefficient (ICC) is a relative index of reliability that normalizes measurement error relative to the heterogeneity of the participants. The ICC can vary between 0 and 1.0 and interpretation can be as follows: < 0.40 *poor* agreement; 0.40 to 0.59 *fair* agreement; 0.60 to 0.74 *good* agreement; 0.75 to 1.00 *excellent* agreement (Cicchetti, 2001).

- The technical error of measurement (TEM) is a measure of imprecision and is defined as the square root of measurement error variance. It is adopted by the International Society for the Advancement of Kinanthropometry (ISAK) as the most common way to express error in anthropometry. As there is a positive association between TEM and measurement size (large mean values of measurement are associated with high TEM and small ones with low TEM), it is not recommended that TEMs are compared directly. Instead, a measure of the coefficient of variation of TEM, the relative TEM, is used to facilitate comparisons between different measures or indices. Interpretation is dependent on the analysed method and for that reason there are no exact cutoffs for acceptance but rather published measurement-specific indications of the precision required for a reliable measurement (Ulijaszek & Kerr, 1999).
- The coefficient of reliability (R) reflects how much of the between-participant variance is free from measurement error. It ranges from 0 to 1 and although there are no recommended values for R, Ulijaszek and Kerr (1999) suggest that a cut-off of 0.95 be used (i.e. a human measurement error of up to 5%).
- The typical error (TE) describes the error in interpreting an individual's test score (i.e. it quantifies the precision of individual scores on a test). The lower the reliability, the greater the TE, and the less precise the scale (Hopkins, 2000), however, it is still the researcher's responsibility to decide whether the identified difference impacts the conclusions drawn from these measurements.

3.17.1 Male participants

All measurements have excellent agreement according to the ICC (between 0.75 and 1.00). The coefficient of reliability is above 0.95 for every measure except waist-to-hip

ratio which has a 0.8 value. Despite being below the suggested cut-off value of 0.95, this value is not an indicator of poor reliability but rather a limitation of this coefficient. This is the reason ICC and R should not be reported in isolation without either the TEM or TE because they are known to have a relationship with between-subjects variability (e.g. low between-subjects variability can reduce the coefficients even when trial-to-trial variability is low) (Weir, 2005). In this case we can see that the standard deviations of both tests are very low (0.03) which can explain this drop in the reliability coefficient while in the presence of low TEM and TE. Moreover, in this study our skinfolds TEM (%) are all within the accepted values for accredited anthropometrists (< 7.5% for beginner anthropometrist; < 5.0% for skilled anthropometrist) (Gore *et al.* 1996) meaning that there is a low degree of error involved in these measurements. In summary, these results support the good reproducibility of these measurements in male participants.

Table 3.2 Reliability of measurements of physical characteristics in male participants

Variables	M ± SD (1st test)	M ± SD (2nd test)	ICC	TEM (%)	R	TE (%)
Stature (cm)	180 ± 9	179 ± 9	1.00	0.1	1.00	0.1
Body mass (kg)	72.9 ± 9.3	73.1 ± 9.5	1.00	0.6	1.00	0.5
Waist-to-hip ratio	0.83 ± 0.03	0.83 ± 0.03	0.85	1.7	0.80	1.5
Bicep skinfold (mm)	3 ± 1	3 ± 1	0.92	2.6	0.98	5.3
Tricep skinfold (mm)	9 ± 2	9 ± 2	0.94	0.5	1.00	5.3
Sub-scapular skinfold (mm)	10 ± 2	10 ± 1	0.96	0.7	1.00	2.9
Suprailiac skinfold (mm)	14 ± 4	14 ± 4	0.98	1.9	0.99	4.1
Sum of skinfolds (mm)	36 ± 6	36 ± 6	0.98	0.4	1.00	2.2
Body fat (%) - skinfolds	16.5 ± 2.5	16.4 ± 2.4	0.98	0.2	1.00	2.1
Fat-free mass (kg) - BIA	66.5 ± 7.8	66.4 ± 8.2	0.99	0.2	1.00	0.7
SMM (kg) - BIA	37.6 ± 4.7	37.8 ± 5.0	0.99	0.7	1.00	0.5
Body fat (kg) - BIA	20.6 ± 14.9	20.5 ± 14.6	1.00	0.3	1.00	0.7
Body fat (%) - BIA	22.0 ± 11.6	21.8 ± 11.6	1.00	1.3	1.00	0.7
VFA (cm^2) - BIA	119.9 ± 71.6	121.6 ± 69.6	1.00	2.9	1.00	4.3
$V\dot{O}_{2max}$ (ml/min)	3055 ± 478	3072 ± 438	0.93	1.2	0.99	3.8
$V\dot{O}_{2max}$ (ml/kg/min)	41.9 ± 3.1	42.1 ± 4.0	0.75	1.1	0.98	4.2
Maximum heart rate (bpm)	182 ± 15	182 ± 15	0.99	0.5	1.00	0.9

N=10; SMM = Skeletal muscle mass; VFA = Visceral fat area; $V\dot{O}_{2max}$ = maximal oxygen consumption;

Note: In this study the variables obtained through bioelectrical impedance analysis (BIA) were assessed in a different sample of 10 male participants as the equipment (Inbody 720) was only purchased by the University after the male reliability study was

already completed. For this reason 10 male participants were recruited for the sole purpose of assessing the reliability of the Inbody 720 measures.

3.17.2 Female participants

All measurements, except maximum heart rate, have excellent agreement according to the ICC (between 0.75 and 1.00). The coefficient of reliability for most measures is above 0.95, however, three skinfolds (bicep, tricep and subscapular) and maximum heart rate have values below this suggested cut-off point. As in the previous male study, the ICC and R of one of these measurements (maximum heart rate) are affected by the very low standard deviations (6 and 5 for the first and second test respectively) because the respective TEM and TE values remain low. Despite no apparent reliability problems with most measures, the TEM and TE values for the skinfolds are higher than expected. The sub-scapular and suprailiac skinfolds are of special concern as they have a reported error of 12% which is well above the accepted levels for accredited anthropometrists (< 7.5% for beginner anthropometrist; < 5.0% for skilled anthropometrist) (Gore *et al.* 1996). A possible explanation of the differences in the amount of error present in this study and the male study skinfolds is that in this sample there was a higher fat accumulation on one of the sites (suprailliac), which might have led to an increased difficulty to measure skinfold thickness and therefore more frequent measurement errors. Additionally, the access to the sub-scapular skinfold site was made difficult in female participants due to the restriction of sports clothing. In summary, most measurements are reproducible with a low amount of error, however, due to the higher amount of error reported in the skinfolds measures, this technique was replaced by bioelectrical impedance in the main studies of this thesis with female participants.

Table 3.3 Reliability of measurements of physical characteristics in female participants

Variables	M ± SD (1^{st} test)	M ± SD (2^{nd} test)	ICC	TEM (%)	R	TE (%)
Stature (cm)	166 ± 7	166 ± 7	1.00	0.3	1.00	0.3
Body mass (kg)	62.2 ± 10.3	62.5 ± 10.4	0.99	1.1	1.00	1.2
Waist-to-hip ratio	0.74 ± 0.05	0.74 ± 0.05	0.91	0.1	1.00	1.9
Bicep skinfold (mm)	8 ± 2	8 ± 2	0.91	7.9	0.93	9.2
Tricep skinfold (mm)	16 ± 4	17 ± 4	0.91	5.9	0.94	7.8
Sub-scapular skinfold (mm)	12 ± 4	13 ± 4	0.97	11.9	0.88	6.1
Suprailiac skinfold (mm)	20 ± 12	19 ± 10	0.94	12.0	0.95	13.1
Sum of skinfolds (mm)	56 ± 18	56 ± 18	0.97	1.2	1.00	5.4
Body fat (%) - skinfolds	29.5 ± 4.1	29.6 ± 4.0	0.97	0.8	1.00	2.5
Fat-free mass (kg) - BIA	46.3 ± 5.8	46.8 ± 6.0	0.98	2.2	0.97	2.0
SMM (kg) - BIA	25.7 ± 3.4	26.0 ± 3.5	0.98	2.6	0.96	2.0
Body fat (kg) - BIA	15.9 ± 5.1	15.8 ± 5.2	0.98	2.1	1.00	4.3
Body fat (%) - BIA	25.1 ± 4.1	24.7 ± 4.6	0.93	3.6	0.96	4.6
VFA (cm^2) - BIA	51.5 ± 20.0	51.2 ± 20.4	0.97	1.1	1.00	6.4
$\dot{V}O_{2max}$ (ml/min)	2132 ± 356	2131 ± 371	0.99	0.1	1.00	1.7
$\dot{V}O_{2max}$ (ml/kg/min)	34.4 ± 4.0	34.1 ± 3.9	0.97	2.2	0.97	2.0
Maximum heart rate (bpm)	192 ± 6	191 ± 5	0.49	1.3	0.72	2.0

N=10; SMM = Skeletal muscle mass; VFA = Visceral fat area; $\dot{V}O_{2max}$ = maximal oxygen consumption;

Chapter 4: Effects of an acute bout of aerobic exercise on immediate and subsequent three-day food intake and energy expenditure in active and inactive men (Study 1)

4.1 Introduction

Exercise is well-recognised as a means to maintain or induce decreases in body mass but the extent to which it beneficially affects appetite control is not clear (Martins *et al.,* 2008). Moreover, the efficacy of exercise in decreasing body mass varies between individuals so there is a need to identify behavioural and physiological compensatory responses that account for these differences (Blundell *et al.,* 2003; King *et al.,* 2007) and ascertain if these compensatory responses differ according to how much physical activity is habitually undertaken (Martins *et al.,* 2008).

Several studies have investigated the relationship between individuals' physical activity and the ability to control energy intake in the short-term using the preload-test meal paradigm (Long *et al.,* 2002; Van Walleghen *et al.,* 2007). Long and colleagues (2002) compared this effect in male regular exercisers and non-exercisers, and suggested that regular exercisers had an increased accuracy of short-term control of food intake compared with non-exercisers. Additionally, Van Walleghen *et al.* (2007) reported that the short-term energy control (i.e. over the course of the testing day) was more accurate in active compared to inactive adults, independent of the age supporting the beneficial role of exercise on the short-term control of appetite. Findings from a 6-week

intervention study (Martins *et al.,* 2007b) also support previous cross-sectional findings and suggest that exercise may lead to a more sensitive response to previous energy intake.

Notwithstanding the evidence that studies using the preload-test meal paradigm can provide regarding the relationship between physical activity status and the ability to control energy intake in the short-term, their findings are specific to situations where energy intake is manipulated. Therefore, to better understand the relationship between physical activity and appetite there is a need to investigate the effects of exercise-induced energy deficits on energy intake.

Several studies have investigated the effects of an acute bout of exercise on food intake and hunger in men with varying findings. Hunger does not change (Imbeault *et al.,* 1997, King *et al.,* 1997, King *et al.,* 2010b), or transiently decreases (Broom *et al.,* 2007, Broom *et al.,* 2009, King *et al.,* 2010a, Martins *et al.,* 2007b, Thompson *et al.,* 1988, Westerterp-Plantenga *et al.,* 1997) after an acute bout of exercise. Furthermore changes in hunger might not influence food intake as most studies reported no effects of an acute bout of exercise on subsequent energy intake (Imbeault *et al.,* 1997, King *et al.,* 2010ab, Thompson *et al.,* 1988). Fewer studies have reported decreases (Westerterp-Plantenga *et al.,* 1997, Jokisch *et al.,* 2012) and increases in energy intake after an exercise bout (Martins *et al.,* 2007b, Verger *et al.,* 1994) but when adjusting for the energy expended during exercise, one of these studies (Martins *et al.,* 2007b) found a decrease in relative energy intake.

Despite the need to investigate effects of exercise on food intake in the inactive, overweight and obese, most studies have examined only regular exercisers (Imbeault *et al.,* 1997, King *et al.,* 1997, King *et al.,* 2010b, Verger *et al.,* 1994). Studies that have examined inactive-to-moderately-active individuals, reported no change (Harris & George, 2008, Klausen *et al.,* 1999), an increase (Martins *et al.,* 2007b) or a decrease (Westerterp-Plantenga *et al.,* 1997, Jokisch, *et al.,* 2012) in food intake after exercise. Furthermore, several limitations apply to these studies such as the use of *ad libitum* buffet-style meals (Blundell *et al.,* 2010) and the estimation of energy expenditure, through the compendium of physical activity (Ainsworth *et al.,* 2000) and American College of Sports Medicine (Glass & Dwyer, 2007). Moreover, some studies did not measure energy expenditure (Westerterp-Plantenga *et al.,* 1997, Harris & George, 2008) and used short observation periods (within the same day) (Martins *et al.,* 2007b, Westerterp-Plantenga *et al.,* 1997, Jokisch *et al.,* 2012, Harris & George, 2008) that might not have been long enough to detect delayed responses in the variables assessed. To overcome some of these limitations the study presented in this chapter increased the observation period and used different methods to measure *ad libitum* energy intake in the laboratory (Sussex Ingestion Pattern Monitor - SIPM) and free-living energy expenditure (Actiheart).

To the author's knowledge, no study has examined the effects of an acute bout of moderate-intensity exercise on immediate and subsequent three-day energy intake and energy expenditure in active and inactive men. Nevertheless, previous research has suggested that exercise leads to a more sensitive response to previous energy intake (Martins *et al.,* 2007b) and have a beneficial influence on appetite-regulating hormones (Stensel, 2011). Therefore, it was hypothesised that active individuals would

demonstrate an improved acute control of energy intake by compensating for the exercise-induced energy expenditure better than inactive individuals. A better understanding of how exercise affects appetite, feeding behaviour and physical activity energy expenditure during the hours and days following exercise might provide insight into how to successfully manage possible compensatory responses.

4.2 Methods

4.2.1 Participants

This study was advertised as examining the relationship between food, activity and mood. Sixty-nine participants requested more information about the study from which forty-four were invited for a preliminary visit. Thirty-five participants met the inclusion criteria for the study but five participants withdrew from the study. Two of these participants were not willing to follow protocol, two withdrew due to facilities/equipment problems and one chose not to state a reason. A consort diagram of this study is represented in figure 4.1.

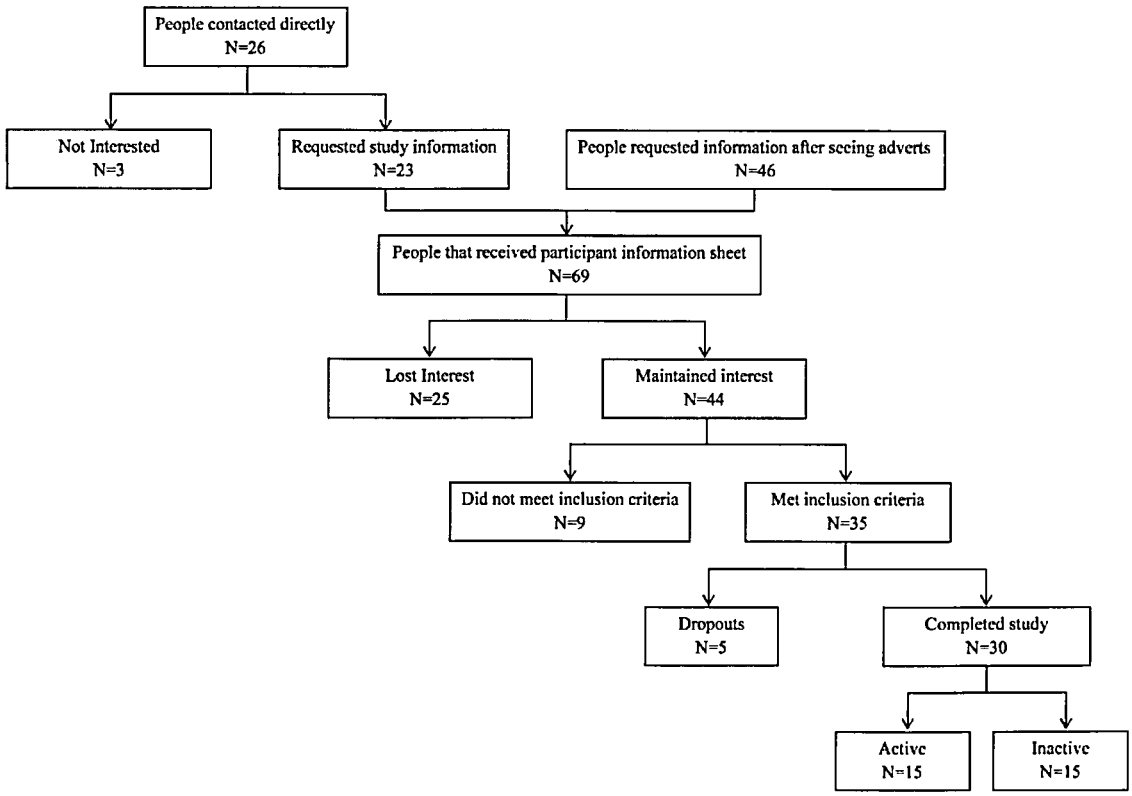


Figure 4.1 Consort diagram of the study.

Participants were recruited according to the following inclusion criteria:

- Male aged 18-35 years;
- Normal or Overweight (body mass index between 18.5 and 29.9 kg/m);
- Healthy (no known chronic diseases);
- Non-smokers;
- Not dieting;
- Body mass stable (±2 kg) for 6 months prior to the study;
- Not being highly restrained eaters (scoring less than 18 or 66% in the cognitive restraint scale of the revised version of the Three Factor Eating Questionnaire, TFEQ-R18);
- Not disliking or having allergies to the foods provided;
- Not taking medications that could affect food intake or metabolism.

Participants mean score for cognitive restraint based on the revised version of the Three-Factor Eating Questionnaire (Karlsson *et al.,* 2000) was 10.5 ± 3.2 (25.2 ± 17.8%) for the active and 10.7 ± 2.8 (25.9 ± 15.5%) for the inactive group. Self-reported weekly physical activity assessed by a modified version of Godin Leisure-Time Exercise Questionnaire (GLTEQ) (Godin & Shepard, 1985) was used to allocate participants to the active (engaged in regular exercise and undertaking at least 150 minutes of moderate-intensity physical activity per week) and inactive groups (did not engage in regular exercise and did not meet the minimum physical activity recommendation guidelines of 150 minutes of moderate-intensity physical activity per week) (Department of Health, 2004). Veracity of self-reported measures of physical activity was confirmed with a posteriori analysis of the Actiheart data that calculated

individual Physical Activity Level (PAL) by dividing participants' total energy expenditure in a 24-hour period by their basal metabolic rate. The active group had a mean daily physical activity level of 1.80 ± 0.19 and the inactive 1.54 ± 0.13, which according to the classification of lifestyles in relation to PAL in adults (WHO, 2004) would identify them as having an active to moderately active lifestyle (1.70-1.99) and a sedentary to light activity lifestyle (1.40-1.69), respectively.

4.2.2 Study design

All participants were blinded to the true purpose of the study to minimise participant-expectancy effects and were informed that the purpose of the investigation was to assess how food and physical activity affected mood. Approximately a week before the experimental days, participants attended the laboratory for one preliminary session consisting of two exercise tests (submaximal and maximal cycling tests), screening and habituation with all procedures as described in chapter 3. After the preliminary session, participants were allocated to either the active or inactive group according to their habitual physical activity and started a randomised crossover study with exactly seven days between both conditions (exercise and control). On the experimental days, participants arrived at the laboratory between 8:30 and 9:30 am after fasting overnight for 10 hours (only water consumption was permitted). On arrival participants consumed a standard breakfast within 15 minutes (Figure 4.2). On the exercise experimental day, participants rested for two hours and cycled for one hour at 50% of maximum oxygen uptake with the rest periods before and after the 60 minute exercise period. On the control experimental day this was equivalent to three hours of rest from the end of breakfast until the beginning of lunch. From the end of breakfast until the beginning of lunch participants could drink water *ad libitum*. After consuming the *ad libitum* lunch,

participants were fitted with an Actiheart and given a food diary that was used to estimate food intake and energy expenditure for the remainder of the experimental day and subsequent three days. At the end of the study participants were debriefed about the true purpose of the study.

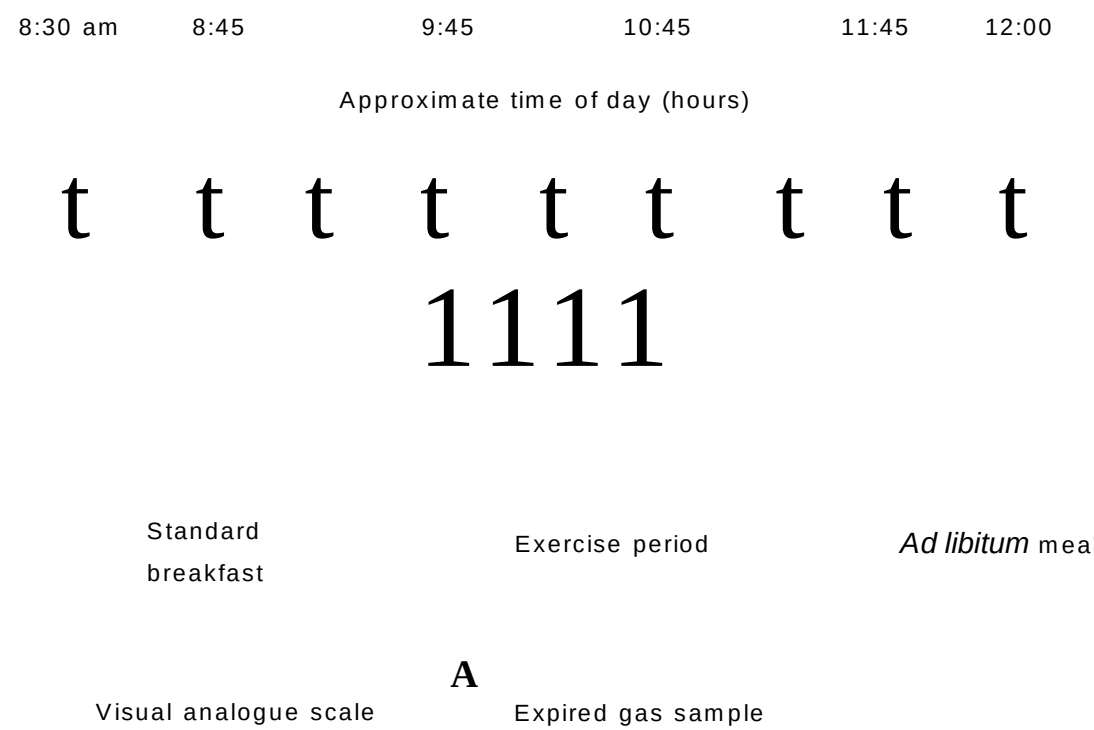


Figure 4.2 Schematic representation of the laboratory period of the experimental days.

4.2.3 Subjective ratings of hunger

During the experimental days, hunger ratings were assessed with paper-based 100-mm visual analogue scales (VAS) before and after breakfast, and at 30 min intervals thereafter until the end of lunch (Figure 4.2). The VAS was preceded by the question

"how hungry do you feel?" anchored on the left by "not at all hungry" and on the right by "very hungry" (Flint *et al.*, 2000). Participants placed a vertical mark through the line at the point which best matched their present feeling of hunger. The distance from the left anchor to the vertical mark was then used as the hunger score.

4.2.4 Laboratory energy expenditure

Expired air samples were collected in 150 l Douglas Bags (Harvard Apparatus, Edenbridge, Kent, UK) at 15 minutes intervals during the 60 minute exercise and rest period of the experimental days (Figure 4.2). Samples were analysed using an oxygen/carbon dioxide gas analyser (Dual Gas Analyser GIR250, Hitech Instruments, Luton, UK) and a dry gas meter (Harvard Apparatus, Edenbridge, Kent, UK) determined expired air volumes that were corrected to standard temperature, pressure and dry gas. This ensured that participants cycled at 50% of their $V\dot{O}_{2max}$ and estimated energy expenditure by indirect calorimetry (Frayn, 1983).

4.2.5 Laboratory energy intake

On each experimental day, participants ate their breakfast and *ad libitum* lunch alone in individual air-conditioned testing cubicles equipped with Sussex Ingestion Pattern Monitors (SIPM). During lunch, participants were instructed to “eat as much or as little as they wanted” and food intake (in grams) was covertly monitored using the SIPM. To ensure participants did not use the empty plate as an external cue to end their meal, the SIPM was programmed to prompt the participant to call the experimenter, using a call button, once at least 300 g were consumed to receive a refill. This process was repeated until the participants indicated that they had finished eating.

4.2.6 Free-living energy intake

Participants were instructed to weigh and record all items of food and drink consumed both at home and outside the home in food diaries for the remainder of the experimental days and subsequent three days. All participants received guidance on how to complete the dietary record and measure food portions. When weighing was not possible, participants were asked to estimate portion sizes using standard household measures. Immediately upon receipt, food diaries were reviewed in the presence of the participant to ensure completeness and legibility, with any missing or unclear items being corrected. Food diaries were analysed to estimate energy and macronutrient intake using the dietary analysis software NetWisp (version 3.0; Tinuviel Software, Warrington, UK) with unlisted foods being inputted according to their nutritional label.

4.2.7 Free-living energy expenditure

Free-living energy expenditure for the remainder of the experimental day and subsequent three days was estimated using an Actiheart (Cambridge Neurotechnology, Cambridge, UK) as described in section 3.15. Participants were told to wear the monitor at all times, when awake or asleep including when washing or swimming. At the end of the three-day period, participants returned the Actihearts and the data were downloaded using a docking station and analysed using its commercial software.

4.2.8 Percentage of energy compensation

Percentage of energy compensation was calculated for the *ad libitum* lunch meal, and for each one of the daily energy intakes (i.e. experimental day and subsequent 3 days) based on a formula used in previous studies (Jokisch *et al.*, 2012; Pomerleau *et al.*,

2004). To calculate the percentages compensation for the *ad libitum* lunch meals and for each day the following formulas were applied:

[(lunch energy intake in exercise condition - lunch energy intake in control condition) / (net exercise-induced energy expenditure)] x 100

[(energy intake of day A in exercise condition - energy intake of day A in control condition) / (net exercise-induced energy expenditure)] x 100

In the latter, "A" denotes the day for which the percentage compensation is being calculated.

When positive, the percentage compensation values indicate that over the analysed period of time, energy intake was greater in the exercise than in the control condition while negative values indicate a greater intake in the control than in the exercise condition. A value of 100% indicate complete compensation of the net exercise-induced energy expenditure (i.e. the excess energy intake at the exercise compared with the control condition matched the net exercise-induced energy expenditure). A value of 0% indicates no compensation (i.e. energy intake was the same in both conditions).

4.2.9 Statistical analyses

Sample size was estimated with the nQuery Advisor software (nQuery Advisor 6.01, Statistical Solutions, Cork, Ireland). On the basis of previous studies with men participants (Harris & George 2008; Martins *et al.*, 2007a; Verger *et al.*, 1994; Westerterp-Platenga *et al.*, 1997) this study aimed to detect a mean difference of energy

intake at the first meal after the exercise compared with the control conditions of 930 kJ (standard deviation of 910 kJ). With an alpha of 5% for statistical significance and statistical power set at 80%, the estimated sample size was 15 participants per group.

Data were analysed using the Statistical Package for the Social Sciences software for windows (SPSS 19.0, Chicago, IL, U.S.A.). Differences between groups for baseline characteristics, relative intensity of exercise (% of $\dot{V}O_{2max}$), ratings of perceived exertion (RPE) during exercise and the net exercise-induced energy expenditure were assessed by independent Student's t-tests. Percentages of energy compensation were compared between groups using a one-way ANOVA with the Welch test (when homogeneity of variance was violated). Two-way mixed-model ANOVAs (Group × Condition) compared the experimental day's lunch energy intake, energy expenditure, heart rate and respiratory exchange ratio (RER). Three-way mixed-model ANOVAs (Group × Condition × Time) compared subjective hunger ratings, daily energy expenditure and daily energy and macronutrient intakes. In these analyses energy intake on the experimental day was calculated by summing participants' energy intake throughout the day (breakfast + *ad libitum* lunch + remainder of experimental day). However, the same formula could not be applied to macronutrient intake because the macronutrient values for breakfast and lunch of the experimental day were fixed. Therefore, macronutrient intake for the experimental day is limited to the free-living period of that day (i.e. remainder of the experimental day). Where appropriate, post hoc tests were performed using Bonferroni adjustments. Cohen's *d* (standardised mean difference) effect sizes were calculated by dividing the difference between means by the pooled standard deviation thus reflecting differences expressed in standard deviation units. According to Cohen's (1988) guidelines, effect sizes may be conservatively interpreted as small (0.2), medium (0.5), and large (0.8) effects. In addition, 95% confidence intervals were

determined for energy intake, macronutrient intake, energy expenditure and percentage of energy compensation. Means and standard deviations (mean ± SD) are presented for all outcomes unless otherwise stated. Statistical significance was accepted at the 5% level.

4.3 Results

4.3.1 Baseline characteristics

Participants' baseline characteristics are presented in table 4.1. Active participants had greater VC^max and lower BMI, percentage of body fat and waist-to-hip ratio than inactive participants *(p* < 0.05). There were no differences in age, stature and body mass *(p* > 0.05).

Table 4.1 Participants baseline characteristics

	Active	**Inactive**
Age (years)	22.5 ±4.3	23.8 ±3.2
Stature (m)	1.79 ±0.07	1.77 ±0.06
Body mass (kg)	73.1 ±9.7	78.8 ±8.2
BMI (kg-nf2) *	22.6 ±2.0	25.1 ±2.4
Waist-to-hip ratio **	0.81 ±0.02	0.86 ± 0.04
Body fat (%) **	14.3 ±3.4	22.2 ±3.8
VOimax (ml kg 1 min'1) **	44.6 ±5.0	35.5 ±5.2

N=15 per group; values presented as mean ± SD.

BMI = body mass index; V02max = maximal oxygen consumption.

* Means significantly different *(p* < 0.05).

** Means significantly different *(p* < 0.01).

4.3.2 Exercise responses and energy expenditure on the experimental days

Relative intensity of exercise, RPE and RER did not differ between active and inactive groups during exercise (51.0 ± 3.8% vs. 51.4 ± 3.4% of $\dot{V}O_{2max}$; 11.3 ± 1.3 vs. 12.1 ± 1.0; and 0.88 ± 0.09 vs. 0.93 ± 0.06; $p > 0.05$). Heart rate differed between groups during the control experimental day (active 57 ± 7 bpm vs. inactive 65 ± 8 bpm, $p = 0.005$, $d = -1.15$) but not during the exercise experimental day (active 109 ± 9 bpm vs. inactive 114 ± 13 bpm, $p = 0.21$, $d = -0.48$). There was a condition ($p < 0.001$, $d = 7.69$) and group ($p = 0.045$, $d = 0.14$) effect and a group x condition interaction ($p = 0.002$, $d = 1.26$) for energy expenditure during the 60 minutes of exercise and equivalent control period. Post hoc analyses demonstrated that active participants expended more energy than inactive participants during exercise (mean difference = 231 kJ; 95% CI 45 to 417 kJ, $p = 0.013$, $d = 1.0$) but not during the equivalent resting period (mean difference = 27 kJ, 95% CI -83 to 30 kJ, $p = 0.33$, $d = -0.38$). Net exercise-induced energy expenditure was greater in active than inactive participants (1579 ± 61 kJ vs. 1322 ± 48 kJ; $p = 0.003$, $d = 1.26$). During the remainder of the experimental days there was a group effect for energy expenditure (active vs. inactive: 7994 ± 2924 kJ vs. 6344 ± 1220 kJ, $p = 0.024$, $d = 0.76$) and physical activity energy expenditure (active vs. inactive: 3344 ± 2553 kJ vs. 1911 ± 1095 kJ, $p = 0.011$, $d = 0.76$) but there were no other main or interaction effects ($p > 0.05$).

4.3.3 Body mass and water consumption on the experimental days

Body mass and water consumption during the laboratory period of the experimental days are presented in table 4.2. There were condition (exercise vs. control: 76.2 ± 9.4 kg vs. 76.5 ± 9.5 kg; $p = 0.027$, $d = -0.04$) and time (start vs. end: 76.4 ± 9.4 kg vs. 76.3 ±

9.4 kg; p = 0.004, d = -0.01) effects and time x group (p = 0.020, d = -0.89) and condition x time (p = 0.011, d = 0.28) interactions for body mass on the experimental days. However, post hoc tests only revealed a difference in active participants between body mass at the start and end of the 60 minutes cycling/resting period (mean difference = -0.4 ± 0.4 kg; p = 0.001, d = -0.04). There were no condition (p = 0.134, d = 0.25), group (p = 0.481, d = 0.25) or interaction (p = 0.610, d = 0.19) effects for water consumption on the experimental days.

Table 4.2 Body mass and water consumption on the experimental days

	Active		**Inactive**	
	Exercise	Control	Exercise	Control
Body mass (kg) - start	73.1 ± 10	73.5 ± 10.2	79.6 ± 8.1	79.6 ± 8.2
Body mass (kg) - end	72.7 ± 9.9*	73.4 ± 10.2	79.5 ± 8.1	79.6 ± 8.2
Water consumption (mL)	624 ± 287	531 ± 324	533 ± 284	487 ± 249

N=15 per group; values presented as mean ± SD.

* Mean significantly different from the start of exercise (p = 0.001, d = -0.04).

4.3.4 Environmental temperature and humidity

There were no condition (exercise vs. control: 20.9 ± 2.2 °C vs. 20.4 ± 2.4 °C, p = 0.14), group (active vs. inactive: 20.7 ± 2.6 °C vs. 20.6 ± 1.9 °C, p = 0.92) or interaction effects (p = 0.72) for temperature. Likewise there were no condition (exercise vs. control: 35.6 ± 9.0% vs. 35.7 ± 8.4%, p = 0.91), group (active vs. inactive: 36.2 ± 9.0% vs. 35.1 ± 8.4%, p = 0.47) or interaction effects (p = 0.73) for humidity.

4.3.5 Subjective ratings of hunger

There was a main effect of time ($p < 0.001$) for hunger ratings but there were no interactions or other main effects (Figure 4.3). Differences in hunger ratings were also evaluated using AUC values for the time before and after breakfast (08:45-09:00), the following hours until lunch (09:00-12:00), and the time before and after lunch (12:00-12:20). There was a main effect of time ($p < 0.001$) for hunger AUC values but no interactions or other main effects ($p > 0.05$).

Figure 4.3 Subjective feelings of hunger. (n=15 per group; means ± SEM). Hatched rectangles are consumption of meals; dark rectangle is equivalent to the 60 minutes cycling period.

4.3.6 *Ad libitum* lunch energy intake

The energy intake at the *ad libitum* lunch meal for active and inactive participants on both experimental days is presented in table 4.3. There were no main effects or interactions for absolute energy intake at lunch ($p > 0.05$), however, there was trend with a moderate effect size for the inactive group's absolute energy intake at lunch to be higher on the exercise day compared to the control day (mean difference = 432 kJ; 95% CI -46.5 to 910.7 kJ, $p = 0.073$, $d = 0.42$). After adjustment of absolute EI for the energy expended during the 60 min of exercise/rest (relative energy intake, REI), there was a condition effect ($F(1,28) = 26.495$; $p < 0.001$, $d = -0.98$) with a lower REI in the exercise than the control condition (2766 ± 1182 kJ vs. 4007 ± 1385 kJ). Post hoc tests showed REI was lower in the exercise than the control in the active (mean difference = -1596 kJ; 95% CI -2461 to -733 kJ, $p = 0.01$, $d = -1.13$) and inactive (mean difference = -885 kJ; 95% CI -1415 to -355 kJ, $p = 0.03$, $d = -0.84$) groups.

Table 4.3 *Ad libitum* lunch meal energy intake

	Active	Inactive
Absolute EI Exercise condition (kJ)	4712 ± 1364	4577 ± 1126
Absolute EI Control condition (kJ)	4687 ± 1666	4145 ± 1026
Relative EI Exercise condition (kJ)	2680 ± 1229**	2853 ± 1134*
Relative EI Control condition (kJ)	4276 ± 1663	3738 ± 1035

N=15 per group; values presented as mean ± SD; EI = energy intake.

* Means significantly different from control condition ($p = 0.03$, $d = -0.84$).

** Means significantly different from control condition ($p = 0.01$, $d = -1.13$).

4.3.7 Daily energy intake

Daily energy intake for both groups is shown in figure 4.4. One participant in each group did not complete the full four-day food diary, therefore analyses were made with 14 participants per group. There was a time effect ($F(1,26) = 11.422$; $p < 0.001$) but no group, condition or interaction effects ($p > 0.05$) for daily energy intake. Post hoc analyses showed that active participants consumed more energy during the exercise experimental day than the same period of the control condition (mean difference = 2070 kJ; 95% Cl 397 to 3743 kJ, $p = 0.024$, $d = 0.56$). This difference is explained by an increase in energy intake during the remainder (free-living part) of the exercise day compared with the remainder of the control experimental day (mean difference = 1872 kJ; 95% Cl 70 to 3676 kJ, $d = 0.59$). There were no other differences for the active group during the three days after the experimental days ($p > 0.05$). Inactive participants increased their energy intake on the third day after exercise compared with the control (mean difference = 2225 kJ; 95% Cl 414 to 4036 kJ, $p = 0.024$, $d = 0.80$) but there were no differences between other daily energy intakes ($p > 0.05$).

Figure 4.4 Daily energy intake (n=14 per group; means ± SEM); * Means significantly different between conditions $(p < 0.05)$;

4.3.8 Daily macronutrient intake

Daily macronutrient intake for both groups is shown in table 4.4. There was a main effect of time ($p < 0.01$) and a time x group ($p < 0.05$) interaction for the percentage of energy consumed from fat and carbohydrates. Conversely, there were no main or interaction effects for protein intake ($p > 0.05$). Pairwise comparisons showed that fat intake was lower on the experimental day compared to the third day after (27 ± 9% vs. 33 ± 10%, $p = 0.006$, $d = -0.63$) whereas carbohydrate was greater on the experimental day compared to the third day after (59 ± 10% vs. 51 ± 11%, $p = 0.006$, $d = 0.75$). However, post hoc analyses showed that only the active group had a lower fat intake (24 ± 8% to 35 ± 9%, $p = 0.001$, $d = -1.37$) and higher carbohydrate intake (62 ± 9% to 49 ± 10%, $p = 0.004$, $d = 1.39$) on the experimental day compared to the third day after.

Table 4.4 Daily macronutrient intake

		Active			Inactive		
		Protein	Fat	CHO	Protein	Fat	CHO
Exp. day (%)	Ex.	15 ± 5	24 ± 8	61 ± 10	13 ± 4	31 ± 10	56 ± 12
	Con.	14 ± 3	23 ± 7	63 ± 8	13 ± 3	30 ± 10	57 ± 10
Day 1 (%)	Ex.	14 ± 4	30 ± 10	56 ± 12	15 ± 5	36 ± 6	49 ± 6
	Con.	15 ± 5	27 ± 8	58 ± 11	16 ± 2	30 ± 13	54 ± 12
Day 2 (%)	Ex.	15 ± 3	30 ± 9	55 ± 10	15 ± 7	35 ± 9	50 ± 11
	Con.	16 ± 5	29 ± 10	55 ± 12	14 ± 4	33 ± 10	53 ± 10
Day 3 (%)	Ex.	17 ± 5	34 ± 8	49 ± 10	15 ± 5	34 ± 10	51 ± 9
	Con.	16 ± 8	35 ± 10	49 ± 11	15 ± 6	32 ± 13	54 ± 15

N=14 per group; values presented as mean ± SD. Exp. = experimental; Ex. = exercise condition; Con. = control condition; CHO = carbohydrates.

4.3.9 Percentage of energy compensation

Percentages of energy compensation are presented in figure 4.5 and their 95% confidence intervals in figure 4.6. Calculation of percentage of energy compensation at the *ad libitum* lunch showed that active participants did not compensate for the net exercise-induced energy expenditure (14 ± 79%; 95% Cl -32 to 59%) but the inactive group showed some compensation (30 ± 59%; 95% Cl -3 to 63%) (Figure 4.6: panel A). For the entire experimental day, compensation was observed for the active group (127 ± 194%; 95% Cl 16 to 238%) but not for the inactive (-22 ± 288%; 95% Cl -187 to 143%) (Figure 4.6: panel B). Percentage compensation calculated for each of the subsequent two days did not show any compensation for the active (day 1, -18 ± 192%, 95% Cl -128 to 192%; day 2, -44 ± 165%; 95% Cl -139 to 51%) and inactive participants (day 1, -70 ± 465%, 95% Cl -337 to 196%; day 2, -119 ± 444%; 95% Cl -373 to 136%) (Figure 4.6: panels C and D, respectively). Percentage of compensation on the third day after the experimental day showed a compensation for the inactive (172 ± 265%; 95% Cl 20 to 324%) but not for the active group (-17 ± 220%; 95% Cl -143 to 109%) (Figure 4.6: panel E). Group comparisons showed that there were no differences and small effect sizes for the percentages of energy compensation for the *ad libitum* lunch (p = 0.55, d = -0.24) and for day 1 (p = 0.70, d = 0.15) and day 2 (p = 0.56, d = 0.23) after the experimental day. However, there were large effect sizes for the experimental day (p = 0.12, d = 0.67) and day 3 after the experimental day (p = 0.05, d = -0.8). The cumulative percentage of energy compensation over the four days was not different (p = 0.72, d = 0.15) between the active (48 ± 375%; 95% Cl -167 to 264%) and inactive (-39 ± 752%; 95% Cl -470 to 392%) groups (Figure 4.6: panel F).

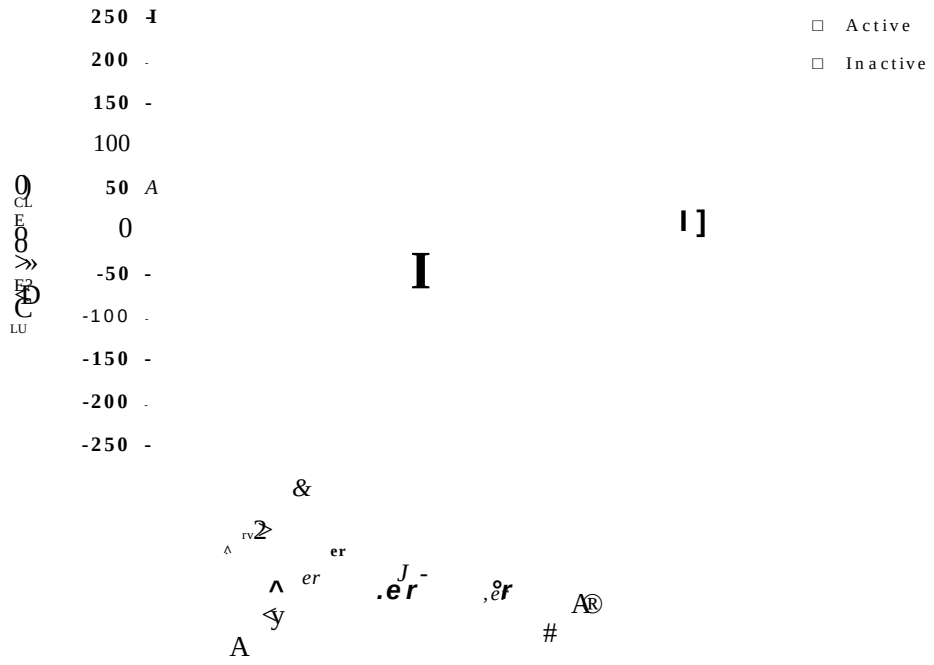


Figure 4.5 Percentages of energy compensation (n=14 per group; means ± SEM); Dashed line indicates complete compensation (100%) of the net exercise-induced energy expenditure.

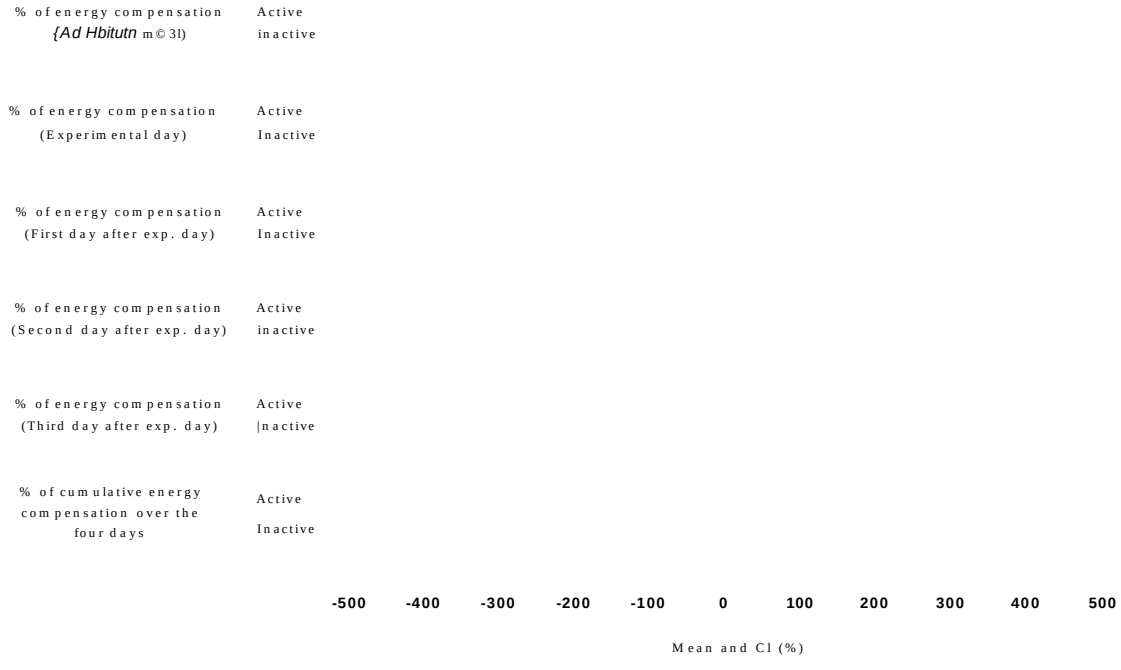


Figure 4.6 Mean percentages of energy compensation and 95% confidence intervals (n=14 per group). Dashed line indicates complete compensation (100%) of the net exercise-induced energy expenditure. Exp. = Experimental.

4.3.10 Daily energy expenditure

Three participants in the active group and one participant in the inactive group did not wear the Actiheart for the third day, therefore analyses are for 12 active and 14 inactive participants. There was only a group effect ($F(1,24) = 5.154$; $p = 0.032$, $d = 0.89$) for total daily energy expenditure, indicating that active participants expended more energy than inactive over the course of the three days (mean difference = 1412 kJ; 95% CI 456 to 2367 kJ). There were no other main effects or interactions ($p > 0.05$) for total daily energy expenditure. Likewise there was only a group effect ($F(1,24) = 12.747$; $p = 0.002$, $d = 1.36$) for physical activity energy expenditure, indicating that the active group was more active than the inactive group (4633 ± 1414 kJ vs. 3039 ± 1019 kJ) over the 3 days after the experimental days (Figure 4.7).

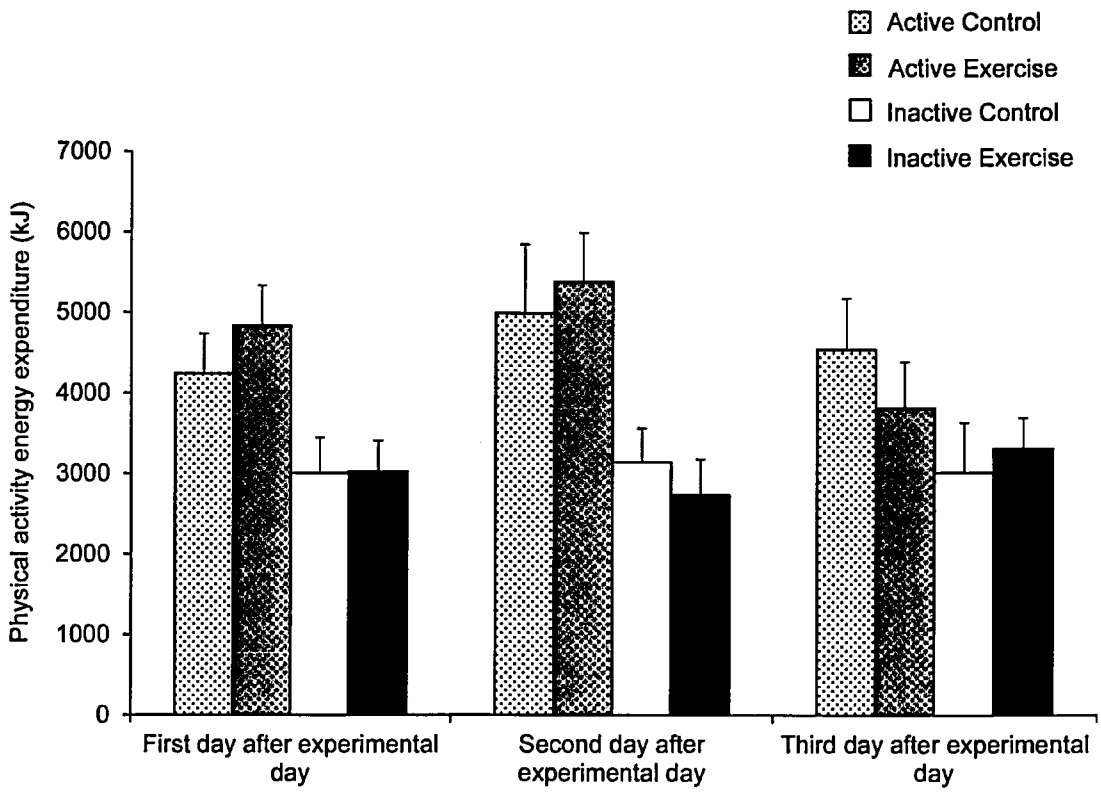


Figure 4.7 Daily physical activity energy expenditure (N=12 active, N=14 inactive; means ± SEM).

4.4 Discussion

To the author's knowledge, the present study is the first to have examined effects of an acute bout of moderate-intensity aerobic exercise on energy intake and expenditure in active and inactive participants over a 4-day period. The novel finding arising from this study is that active participants can immediately increase their energy intake to compensate for exercise-induced energy expenditure on the same day while the inactive participants increased their energy intake only on the third day after exercise. Moreover, both groups maintained their habitual physical activity between conditions suggesting that there were no acute compensatory changes in physical activity.

In the present study, exercise intensity was individually prescribed to reflect fitness status with both groups cycling at approximately the same relative intensity. This adjustment is important since individuals respond differently to an exercise challenge at a fixed absolute intensity (Howley, 2001), and as anticipated, active participants were aerobically fitter which meant they exercised at a greater absolute intensity and therefore expended more energy during exercise than inactive participants. Participants body mass did not change between conditions suggesting that participants remained in energy and fluid balance during the 7 days between the first and the second experimental day. Over the course of the laboratory period of the experimental days there was only a difference between the pre- and post-exercise body mass in the active group (mean difference = -0.4 ± 0.4 kg), which can be explained by the water lost during the exercise period.

Consistent with previous research (Imbeault *et al.*, 1997; Jokisch *et al.*, 2012; King *et al.*, 1997; King *et al.*, 2010b), this study did not find any differences in subjective

hunger ratings either between groups or conditions. A possible explanation for this finding is that previous research has reported only a short-term suppression of hunger during and immediately after vigorous exercise (>60% of $\dot{V}O_{2max}$) (Broom *et al.*, 2007, Broom *et al.*, 2009) whereas moderate-intensity exercise (≈50% of $\dot{V}O_{2max}$) was undertaken in the present study. In addition, energy intake during the *ad libitum* lunch meal did not differ between conditions or groups, again supporting previous research (Harris & George, 2008, Imbeault *et al.*, 1997, King *et al.*, 2010ab, Thompson *et al.*, 1988). However, there was a trend that the inactive group had a greater absolute energy intake at lunch after exercise than control. This finding is reflected in the percentage of energy compensation at the *ad libitum* lunch meal which showed that the inactive group partially compensated for 30% of their net exercise-induced energy expenditure. This result can possibly be explained by inactive participants having a psychological drive to use food as a reward for exercising or the belief that exercise increases appetite (King *et al.*, 2007).

After adjusting for the energy expended during the exercise/rest periods, both groups had a lower lunch REI after exercise than control supporting previous research with active (Imbeault *et al.*, 1997) and inactive individuals (Martins *et al.*, 2007a) demonstrating that exercise induces an acute negative energy balance. In a recent study, with active and inactive participants (Jokisch *et al.*, 2012) habitually active individuals acutely i.e. at the meal immediately after exercise, increased their energy intake whereas the inactive decreased their energy intake. Findings from the present study contradict this work and suggest that at the first meal after exercise, active participants do not change their energy intake and inactive participants potentially increase it. The discrepancy in results between this study and Jokisch *et al.* (2012) study could be

attributable to differences in the study design, such as the methods used to assess energy intake and expenditure, and participant characteristics such as the criteria that defined participants as active and inactive. For instance, Jokisch *et al.* (2012) did not control breakfast, caffeine and alcohol consumption, energy intake was assessed with a buffet-style lunch and energy expenditure was estimated using ACSM metabolic equations.

During the free-living period of this study, the active group increased their food intake during the remainder of the exercise experimental day compared with control, with no other differences observed in the remaining three days, whilst the inactive group only increased their intake on the third day after the exercise day compared with control. Therefore, active individuals were possibly more sensitive in detecting changes in energy balance in the acute phase after exercise, while the inactive participants took longer to detect those changes. A possible explanation for these differences is that active and inactive individuals control their energy intake through deficit thresholds after which compensation occurs. This could be followed by a period where compensation stops until another sufficient energy deficit is created. The higher energy expended during exercise could therefore explain why active compensated within the exercise experimental day and why the inactive only compensated on the third day after two days of increasing their energy deficit. Indeed, considering that the active group is already used to exercising it is possible that its effect is more acute and short-lived than in the inactive group which may require more time to recover from the greater exercise-induced physiological stress.

The calculated percentages of energy compensation for the different time periods in this study confirmed this different timing of compensation between groups and indicated

that the active group compensation in the end of the experimental day was closer to 100% than the one by the inactive group on third day after the experimental day (127 vs. 172%, respectively). These findings suggest that active individuals may not only compensate quicker than inactive but possibly more accurately as well supporting previous research (Long *et al.*, 2002; Martins *et al*, 2007b), which suggests that active individuals have a more sensitive short-term appetite control than the inactive. Moreover, the consistently narrower confidence intervals of the percentages of energy compensation for the active compared with the inactive group are possibly a reflection of the tighter control of energy intake in active participants.

When examining the cumulative percentage of energy compensation over the four days, the active group was only able to compensate for approximately half of their net exercise-induced energy expenditure (48%) whereas the inactive group increased their exercise-induced energy deficit (-39%). From a body mass management perspective, these values would, if sustained over a longer period of time, translate to reductions in body mass. However, this interpretation should be made with caution because whilst the delayed compensation of the inactive group suggests that an exercise-induced effect was still present by the fourth day, the same is not observed with the active group because their full compensation occurred within the exercise experimental day. This result makes the cumulative percentage of energy compensation of the active group difficult to interpret as the exercise-induced effect may have already subsided after the experimental day.

No differences in macronutrient intake were observed between conditions and differences over time were only observed in the active group who consumed less energy

after. As highlighted in a recent review of the effects of exercise on macronutrient intake, previous studies do not show any consistent acute effect of exercise on macronutrient intake and it is therefore possible that these effects are only apparent after a prolonged period of exercise (Elder & Roberts, 2007).

Total energy expenditure and physical activity energy expenditure during the free-living was greater in active than inactive participants, however, there were no differences between conditions suggesting that both groups maintained their habitual physical activity. These results agree with the only study (to the author's knowledge), that has examined the effects of a single bout of exercise on non-exercise physical activity in adults (Alahmadi *et al.,* 2011). In this study, sixteen overweight and obese men performed a single bout of moderate- and high-intensity walking on two separate occasions with non-exercise physical activity being assessed by accelerometers on the three days before each experimental day, on the exercise days and for three days after the experimental days (Alahmadi *et al.,* 2011). This study reported that a single bout of moderate- and high-intensity walking did not alter non-exercise physical activity on the exercise day or on the first 2 days. However on the third day after exercise, an increase was observed after the high- but not after the moderate-intensity exercise session (Alahmadi *et al.,* 2011). These results suggest that higher exercise intensities may be needed to elicit compensatory changes in physical activity. However, it is important to note that the Alahmadi et al. (2011) study did not have a control experimental day and measured non-exercise activity only through accelerometer counts, which does not capture the full energy cost of certain activities such as walking while carrying a load or walking uphill, because acceleration patterns do not change under these conditions (Warren *et al.,* 2010).

walking uphill, because acceleration patterns do not change under these conditions (Warren *et al.*, 2010).

In summary, this study demonstrated that an acute bout of moderate-intensity aerobic exercise did not increase hunger or energy intake at the meal immediately after exercise, but induced an acute compensatory (within the experimental day) increase in energy intake in active men, and a delayed compensatory (third day after the experimental day) increase in energy intake in inactive men. Additionally, these findings occurred in the absence of compensatory changes to daily energy expenditure. Therefore, the results suggest that active individuals compensate for an acute exercise-induced energy deficit quicker than inactive individuals by changing energy intake but not physical activity. These findings provide novel information about the different compensatory responses that active and inactive men have in response to an acute bout of moderate-intensity exercise. Additionally, the cumulative percentage of energy compensation over the four days reinforces the use of exercise as a strategy to induce short-term energy deficits in inactive participants, making it beneficial for individuals aiming to reduce their body mass.

Chapter 5: Effects of an acute bout of aerobic exercise on immediate and subsequent three-day food intake and energy expenditure in active and inactive women (Studies 2 & 3)

5.1 Introduction

As previously discussed, exercise-induced behavioural and physiological compensatory responses in energy intake and/or non-exercise energy expenditure (King *et al.,* 2007) may explain the high inter-variability responses of exercise interventions to reduce body mass. Additionally, these responses may differ according to participants' physical activity (Martins *et al.,* 2008) and sex (Hagobian *et al.,* 2010); therefore, the current chapter follows on the work of study one and examines potential acute exercise-induced compensatory responses in women.

Most studies investigating the effects of an acute bout of exercise on hunger and food intake in active (Finlayson *et al.,* 2009; Hagobian *et al.,* 2012; Lluch *et al.,* 1998; Lluch *et al.,* 2000; Larson-Meyer *et al.,* 2012) and inactive women (George & Morganstein, 2003; Maraki *et al.,* 2005; Reger *et al.,* 1984; Tsofliou *et al.,* 2003; Unick *et al.,* 2010) reported no changes in hunger and/or energy intake. Despite the majority of studies reporting a consistent lack of an acute effect of exercise on energy intake, most of these studies have only assessed energy intake in one subsequent meal one to two hours after exercise (Finlayson *et al.,* 2009; George & Morganstein, 2003; Hagobian *et al.,* 2012; Larson-Meyer *et al.,* 2012; Tsofliou *et al.,* 2003; Unick *et al.,* 2010), so any

compensation that may have occurred later on the day or during subsequent days was not measured.

Several of these studies (George & Morganstein, 2003; Kissileff *et al.,* 1990; Maraki *et al.,* 2005; Reger *et al.,* 1984) have also examined premenopausal women without controlling variables such as the regularity of the menstrual cycles, premenstrual or unusual menstrual symptoms, menstrual phase when testing and the use of hormonal contraceptive preparations. This is a limitation because several studies have reported significantly higher energy intake at the luteal phase than follicular phase and there is evidence that women prone to premenstrual or unusual menstrual symptoms have greater fluctuations on energy intake and appetite (Dye & Blundell, 1997). Additionally mixed findings exist regarding the effects of oral contraceptives (OC) on energy intake with some studies reporting an increase (Eck *et al.,* 1997; Naessen *et al.,* 2007) and others no difference (Bancroft & Rennie, 1993; McVay *et al.,* 2011; Tucci *et al.,* 2010). Other limitations include, the use of *ad libitum* buffet-style meals (George & Morganstein, 2003; Reger *et al.,* 1984, Tsofliou *et al.,* 2003; Unick *et al.,* 2010), the lack of definition of participants' inactivity (Reger *et al.,* 1984; Tsofliou *et al.,* 2003), the estimation of energy expenditure using heart rate equations (George & Morganstein, 2003; Maraki *et al.,* 2005) and the lack of measurement of energy expenditure (Tsofliou *et al.,* 2003).

To the author's knowledge, no studies have examined the acute effects of a single bout of exercise on energy intake and physical activity energy expenditure whilst directly comparing active and inactive women. As in the previous study with men it was hypothesised that active individuals would demonstrate an improved acute control of

energy intake by compensating for the exercise-induced energy expenditure better than inactive individuals. However, due to the potential effects of oral contraceptives (OC) on energy intake (Eck *et al.,* 1997; Naessen *et al.,* 2007) and appetite-regulating hormones in young healthy women (Hirschberg *et al.,* 1996; Karlsson *et al.,* 1992; Naessen *et al.,* 2007) a separate study was conducted to examine women taking oral contraceptives. Therefore, this chapter will be comprised of two studies investigating the effects of an acute bout of exercise on the immediate and subsequent three-day energy intake and expenditure in active and inactive women not using hormonal contraceptives (study two) and taking oral contraceptives (study three). Findings from these studies might provide insight into how to successfully manage possible exercise-induced compensatory responses.

5.2 Methods

5.2.1 Participants

Similarly to study one, these two studies were advertised as examining the relationship between food, activity and mood. One hundred and sixty four participants requested more information about the studies from which seventy-five were invited for a preliminary visit. Twenty-three participants met the inclusion criteria for the second study but seven participants withdrew from this study due to inability to find suitable dates for the experimental days. Similarly, twenty-nine participants met the inclusion criteria for the third study with nine participants withdrawing from this study. From this group of participants, four stated personal reasons, three did not find suitable dates for the experimental days, one did not like breakfast, one did not like to wear the Actiheart and one dropped out due to facilities/equipment problems. A consort diagram of this study is represented in figure 5.1.

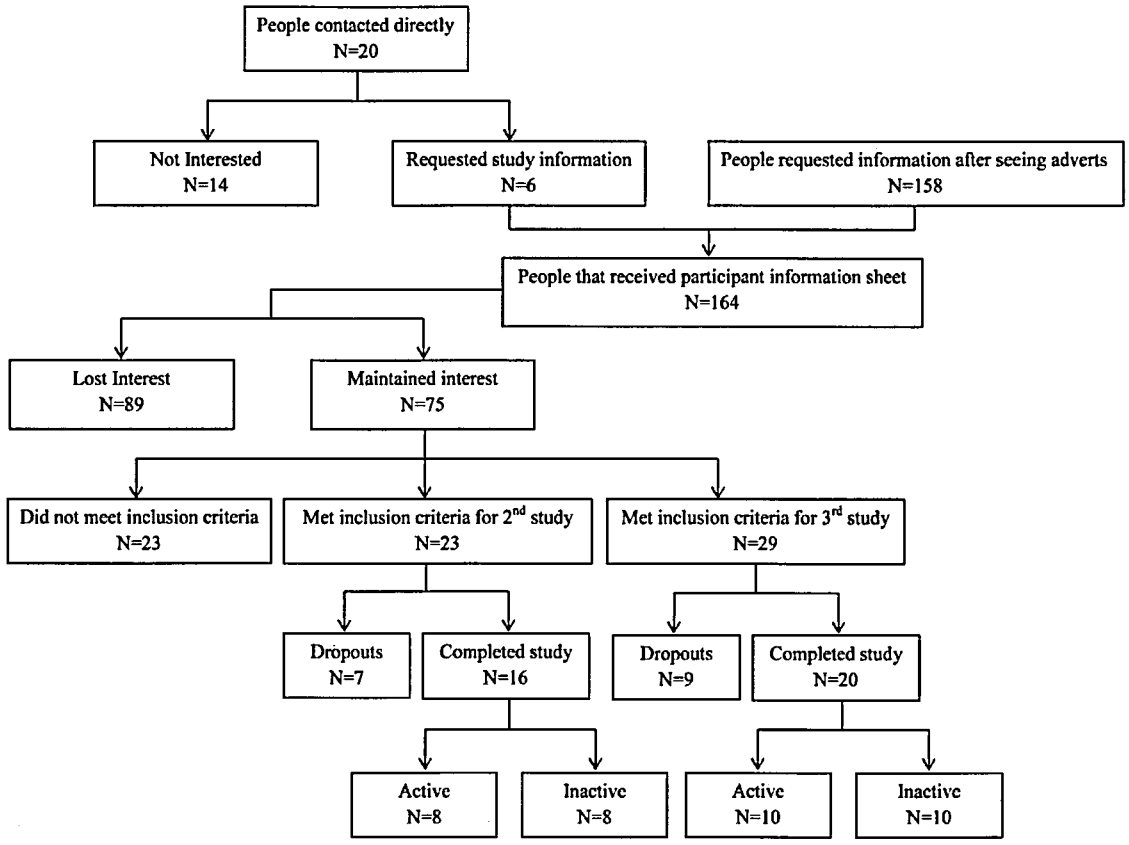


Figure 5.1 Consort diagram of the second and third studies.

The inclusion criteria for these studies were as follows:

- Female aged 18-35 years;
- Normal or Overweight (body mass index between 18.5 and 29.9 kg/m^2);
- Healthy (no known chronic diseases);
- Non-smokers;
- Not dieting;
- Body mass stable (±2 kg) for 6 months prior to the study;
- Not being highly restrained eaters (scoring less than 18 or 66% in the cognitive restraint scale of the revised version of the Three Factor Eating Questionnaire, TFEQ-R18);
- Not disliking or having allergies to foods provided;

- Had regular menstrual cycles (21-35 days);
- Did not suffer from moderate or severe premenstrual symptoms (assessed through the shortened premenstrual assessment form, SPAF);
- Not using hormonal contraceptives (study 2);
- Taking oral contraceptives (study 3; Microgynom 30 [n=6]; Cerazette [n=4]; Yasmin [n=3]; Cilest [n=3]; Femodene [n=1]; Dianette [n=1]; Marvelon [n=1]; Mercillon [n=1])
- Not pregnant or lactating;
- Not taking medications that could affect food intake or metabolism.

Severity of premenstrual symptoms was assessed through the shortened premenstrual assessment form (SPAF; Allen *et al.*, 1991) that consists of 10 items rated on a scale from 1 (not present or no change from usual) to 6 (extreme change, perhaps noticeable even to casual acquaintances). A score greater than 30 indicates moderate premenstrual symptoms (Allen *et al.*, 1991), therefore participants were excluded on this basis. The mean score for the SPAF for the active and inactive groups were 18.1 ± 5.8 and 17.6 ± 5.9 for the second study and 16.8 ± 6.8 and 17.6 ± 5.8 for the third study, respectively.

The mean scores for cognitive restraint based on the revised version of the Three-Factor Eating Questionnaire (Karlsson *et al.*, 2000) for the active and inactive groups were 11.6 ± 3.0 (31.2 ± 16.6%) and 11.0 ± 3.4 (27.8 ± 18.8%) for the second study and 11.6 ± 3.1 (31.3 ± 17.1%) and 10.5 ± 3.3 (25.0 ± 18.5%) for the third study, respectively. Self-reported weekly physical activity assessed by a modified version of Godin Leisure-Time Exercise Questionnaire (GLTEQ) (Godin & Shepard, 1985) was used to allocate participants to the active (engaged in regular exercise and undertaking at least 150

minutes of moderate-intensity physical activity per week) and inactive groups (did not engage in regular exercise and did not meet the minimum physical activity recommendation guidelines of 150 minutes of moderate-intensity physical activity per week) (Department of Health, 2004). Veracity of self-reported measures of physical activity was confirmed with a posteriori analysis of the Actiheart data that calculated individual Physical Activity Level (PAL) by dividing participants' total energy expenditure in a 24-hour period by their basal metabolic rate. The mean daily physical activity level of participants in the second (active: 2.09 ± 0.32; inactive: 1.52 ± 0.15) and third study (active: 1.79 ±0.13; inactive: 1.56 ±0.15) identified the active groups as having an active to moderately active lifestyle (1.70-1.99) and the inactive groups as having a sedentary to light activity lifestyle (1.40-1.69) (WHO, 2004).

5.2.2 Study design

All participants were blinded to the true purpose of the studies (effects of an acute bout of exercise on immediate and subsequent three-day energy intake and expenditure) and were informed that the purpose of the investigation was to assess how food and physical activity affects mood to minimise participant-expectancy effects. Before the experimental days, participants attended the laboratory for one preliminary session consisting of two exercise tests (submaximal and maximal cycling tests), screening and habituation with all procedures as described in chapter 3. After the preliminary session and based on the inclusion criteria, participants were allocated to either the second or third study, and within those to the active or inactive group. Each study was completed in a randomised crossover fashion with approximately 4 weeks (time varied according to participants' menstrual cycle) between both conditions (exercise and control). Randomisation was determined with a randomisation schedule created in excel. In the

second study participants' experimental days were booked during the early to mid-follicular phase (days 5-9) of the menstrual cycle (when oestrogen and progesterone are at their lowest concentrations) (Jonge, 2003) whereas in the third study the experimental days were booked during the first week they restarted taking the oral contraceptives or if continuous when a new pack was started. This control means that findings from these studies are possibly limited to the mechanisms operating at the examined stages, however, this was undertaken to minimise the possible effects of sex hormones in energy intake (Dye & Blundell, 1997) and expenditure (Bowen *et al*., 2011). The experimental days were completed on the same day of the week in both studies to control for dietary and physical activity habits.

On the experimental days, participants arrived at the laboratory between 8.00 and 9.30am after a 10-hour overnight fast with only water consumption permitted. All procedures and measurements thereafter were identical to the ones described in the methods section of the previous chapter.

5.2.3 Statistical analyses

Sample size for the two studies was estimated with the nQuery Advisor software (nQuery Advisor 6.01, Statistical Solutions, Cork, Ireland). On the basis of previous research using female participants (Lluch *et al.,* 2000; Maraki *et al.,* 2005; Martins *et al.,* 2007; Pomerleau *et al.,* 2004) these studies aimed to detect a mean difference of energy intake between the exercise and control conditions of 950 kJ with a standard deviation of 840 kJ. With 5% ($p < 0.05$) as the level of statistical significance and the statistical power set at 80%, the estimated sample size was 13 participants per group in each study.

Data for both studies were analysed using the Statistical Package for the Social Sciences software for windows (SPSS 19.0, Chicago, IL, U.S.A.). Differences between groups for baseline characteristics, relative intensity of exercise (% of V_{O_2}max), ratings of perceived exertion (RPE) during exercise and net exercise-induced energy expenditure were assessed by independent Student's t-tests. Percentages of energy compensation were compared between groups using a one-way ANOVA with the Welch test (when homogeneity of variance was violated). Two-way mixed-model ANOVAs (Group x Condition) compared the experimental day's lunch energy intake, energy expenditure, heart rate and respiratory exchange ratio (RER). Three-way mixed-model ANOVAs (Group x Condition x Time) compared mean daily energy and macronutrient intakes. In these analyses energy intake on the experimental day was calculated by summing participants' energy intake throughout the day (breakfast + ad libitum lunch + remainder of experimental day). However, the same formula could not be applied to macronutrient intake because the macronutrient values for breakfast and lunch of the experimental day were fixed. Therefore, macronutrient intake for the experimental day is limited to the free-living period of that day (i.e. remainder of the experimental day). Subjective hunger ratings and daily energy expenditure were also analysed with three-way mixed model ANOVAs (Group x Condition x Time). Where appropriate, post hoc tests were performed using Bonferroni adjustments. Cohen's d (standardised mean difference) effect sizes were calculated by dividing the difference between means by the pooled standard deviation thus reflecting differences expressed in standard deviation units. According to Cohen's (1988) guidelines, effect sizes may be conservatively interpreted as small (0.2), medium (0.5), and large (0.8) effects. In addition, 95% confidence intervals were determined for energy intake, macronutrient intake, energy expenditure and percentage of energy compensation. Means and standard deviations (mean ± SD)

are presented for all outcomes unless otherwise stated. Statistical significance was accepted at the 5% level.

5.3 Results - Women not using hormonal contraceptives (Study 2)

5.3.1 Baseline characteristics

Participants' baseline characteristics are presented in table 5.1. Active participants have greater $\dot{V}O_{2max}$ and waist-to-hip ratio, and lower percentage of body fat than inactive participants ($p < 0.05$). There were no differences in age, stature, body mass and BMI.

Table 5.1 Participants baseline characteristics

	Active	Inactive
Age (years)	21.9 ± 4.0	24.5 ± 3.5
Stature (m)	1.68 ± 0.07	1.65 ± 0.07
Body mass (kg)	62.1 ± 5.8	62.7 ± 9.9
BMI ($kg \cdot m^{-2}$)	22.2 ± 2.0	23.0 ± 3.1
Waist-to-hip ratio *	0.78 ± 0.03	0.72 ± 0.05
Body fat (%) **	23.6 ± 5.7	32.8 ± 4.2
$\dot{V}O_{2max}$ ($ml \cdot kg^{-1} \cdot min^{-1}$) **	38.8 ± 4.2	26.1 ± 2.3

N=8 per group; values presented as mean ± SD.

BMI = body mass index; $\dot{V}O_{2max}$ = maximal oxygen consumption.

* Means significantly different ($p < 0.05$).

** Means significantly different ($p < 0.01$).

5.3.2 Exercise responses and energy expenditure on the experimental days

Relative intensity of exercise was not different between active and inactive participants during the exercise (50.1 ± 2.1% vs. 55.2 ± 9.5% of $\dot{V}O_{2max}$; $p = 0.17$) but there was a trend for active participants to perceive exercise as less strenuous than inactive participants (RPE: 11.2 ± 1.0 vs. 13.0 ± 2.1; $p = 0.051$). There were no main or interaction effects for RER ($p > 0.05$), however there was a condition ($F(1,14) = 394.2$; $p < 0.001$) and Group ($F(1,14) = 5.2$; $p < 0.05$) effect for heart rate that was different between groups during the control experimental day (active 64 ± 12 bpm vs. inactive 75 ± 7 bpm, $p = 0.021$) but not during the exercise experimental day (active 121 ± 14 bpm vs. inactive 130 ± 10 bpm, $p = 0.12$). There was a condition ($p < 0.001$) and group ($p = 0.01$) effect and a group x condition interaction ($p = 0.014$) for energy expenditure during the 60 minutes of exercise and equivalent resting period. Post hoc analyses demonstrated that active participants expended more energy than inactive participants during exercise (mean difference = 335 kJ; 95% CI 95 to 576 kJ, $p = 0.01$, $d = 1.6$) but not during the equivalent resting period (mean difference = 15 kJ, 95% CI -42 to 72 kJ, $p = 0.59$, $d = 0.29$). Net exercise-induced energy expenditure was greater in active than inactive participants (1137 ± 168 kJ vs. 817 ± 274 kJ; $p = 0.014$, $d = 1.51$). There was a group effect for total energy expenditure (active vs. inactive: 6600 ± 1529 kJ vs. 4625 ± 799 kJ, $p = 0.007$, $d = 1.67$) and physical activity energy expenditure (active vs. inactive: 2970 ± 1386 kJ vs. 1296 ± 512 kJ, $p = 0.006$, $d = 1.65$) during the remainder of the experimental days. There were no other main or interaction effects ($p > 0.05$).

5.3.3 Body mass and water consumption on the experimental days

Body mass and water consumption on the experimental days are presented in table 5.2. There were no group *(p* = 0.79), condition *(p* = 0.26), time *(p* = 0.43) or interaction effects *(p* > 0.05) for body mass on the experimental days. There was a condition *(p* - 0.007, *d* = 0.81) effect for water consumption that was higher during the exercise (604 ± 293 mL) than the control condition (373 ± 299 mL). Post hoc analyses confirmed a difference between conditions in active *{p* = 0.024, *d* = 0.83) but not in inactive participants *(p* = 0.098, *d* = 0.64). There were no other main or interaction effects *(p* > 0.05).

Table 5.2 Body mass and water consumption on the experimental days

	Active		**Inactive**	
	Exercise	Control	Exercise	Control
Body mass (kg) - start	62.6 ±5.3	62.6 ± 5.6	63.6 ± 10.3	63.9 ± 10.7
Body mass (kg) - end	62.5 ±5.3	62.7 ±5.7	63.6 ± 10.2	63.8 ± 10.7
Water consumption (mL)	549 ± 260*	318 ± 333	659 ±348	427 ± 295

N=8 per group; values presented as mean ± SD.

* Mean significantly different from the control condition *(p* = 0.024, *d* = 0.83).

5.3.4 Environmental temperature and humidity

There were no condition (exercise vs. control: 20.3 ± 2.6 ℃ vs. 20.1 ± 2.4 °*C*,*p* = 0.85), group (active vs. inactive: 20.2 ± 2.9 ℃ vs. 20.2 ± 2.0 °*C*,*p* = 1.0) or interaction effects *ip* = 0.81) for temperature. Likewise there were no condition (exercise vs. control: 35.0

± 7.9% vs. 33.8 ± 9.2%, $p = 0.67$), group (active vs. inactive: 34.1 ± 9.0% vs. 34.6 ± 8.1%, $p = 0.89$) or interaction effects ($p = 0.35$) for humidity.

5.3.5 Subjective ratings of hunger

There was a main effect of time ($p < 0.001$) for hunger ratings but there were no interactions or other main effects ($p > 0.05$) (Figure 5.2). Differences in hunger ratings were also evaluated using AUC values for the time before and after breakfast (08:45-09:00), the following hours until lunch (09:00-12:00), and the time before and after lunch (12:00-12:20). There was a main effect of time ($p < 0.001$) for hunger AUC values but no interactions or other main effects ($p > 0.05$).

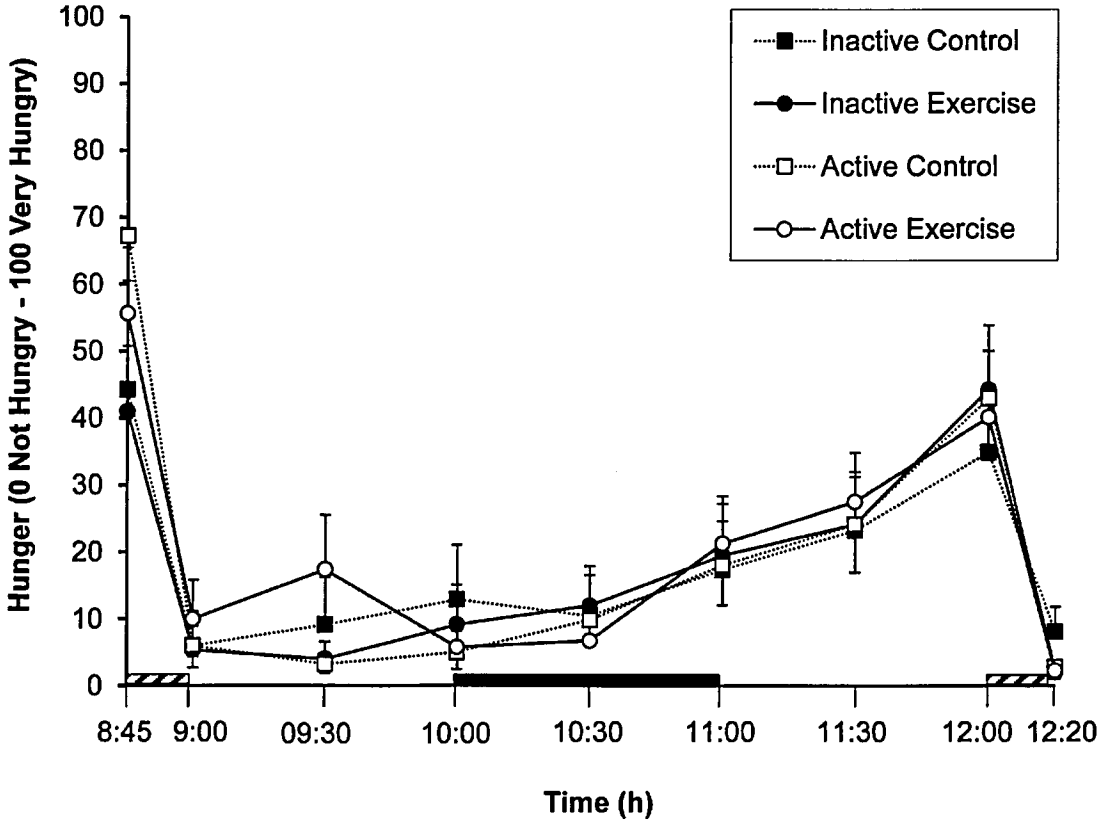


Figure 5.2 Subjective feelings of hunger (n=8 per group; means ± SEM). Hatched rectangles are consumption of meals; dark rectangle is equivalent to the 60 minutes cycling period.

5.3.6 *Ad libitum* lunch energy intake

The energy intake at the *ad libitum* lunch meal for active and inactive participants on both experimental days is presented in table 5.3. There were no main effects or interactions for absolute energy intake ($p > 0.05$). After adjustment of absolute energy intake for the energy expended during the 60 min of exercise/rest (relative energy intake, REI), there was a condition effect ($F(1,14) = 11.735$; $p = 0.004$, $d = -0.79$) with a lower REI in the exercise than the control condition (1417 ± 926 kJ vs. 2120 ± 923 kJ). Post hoc analysis demonstrated a difference in relative energy intake between conditions in the active (mean difference = -1015 kJ; 95% CI -1636 to -394 kJ, $p = 0.01$, $d = -1.28$) but not in the inactive group (mean difference = -392; 95% CI -1016 to 233 kJ, $p = 0.22$, $d = -0.41$).

Table 5.3 *Ad libitum* lunch meal energy intake

	Active	Inactive
Absolute EI Exercise condition (kJ)	2965 ± 583	2458 ± 1296
Absolute EI Control condition (kJ)	2843 ± 1099	2033 ± 619
Relative EI Exercise condition (kJ)	1503 ± 452*	1331 ± 1319
Relative EI Control condition (kJ)	2518 ± 1108	1723 ± 601

N=8 per group; values presented as mean ± SD; EI = energy intake.

* Means significantly different from control condition ($p = 0.01$, $d = -1.28$).

5.3.7 Daily energy intake

Daily energy intake for both groups is shown in figure 5.3. One participant in the inactive group did not complete the full four-day food diary, therefore analyses were made with 8 active and 7 inactive participants. There were no main or interaction effects ($p > 0.05$) for daily energy intake.

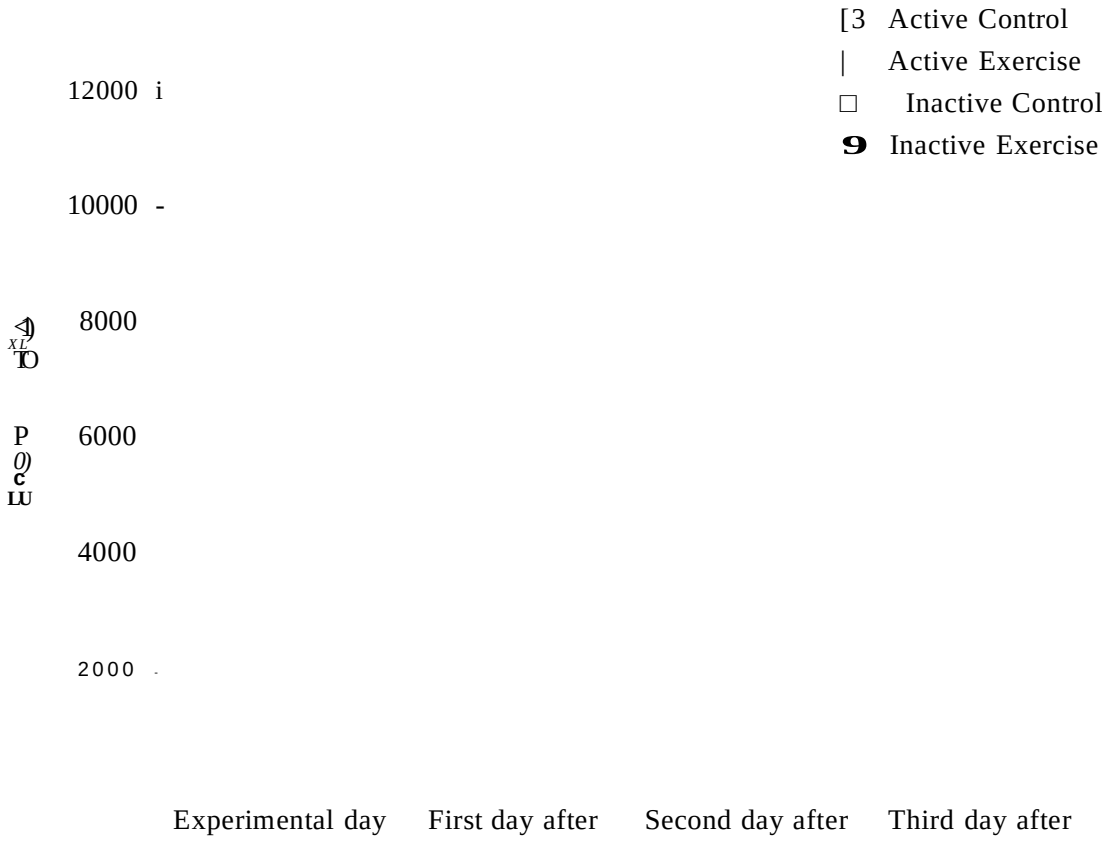


Figure 5.3 Daily energy intake (n=8 for active and n=7 for inactive; means ± SEM).

5.3.8 Daily macronutrient intake

Daily macronutrient intake for both groups is shown in table 5.4. There was a condition effect for the percentage of energy consumed from protein ($p = 0.016$, $d = -0.72$) and carbohydrates ($p = 0.031$, $d = 0.69$) suggesting that participants consumed more

carbohydrates and less protein on the exercise than the control condition (CHO: 53 ± 5% vs. 49 ± 7%; Protein: 14 ± 3% vs. 16 ± 4%). Conversely, there were no main or interaction effects for fat intake ($p > 0.05$). Post hoc analysis demonstrated that only the inactive group consumed less protein during the exercise than the control condition (mean difference = -3%; 95% CI -4 to -1%, $p = 0.005$, $d = -1.00$) and a trend for higher carbohydrate consumption during the exercise compared to the control condition (mean difference = 5%; 95% CI 0 to 10%, $p = 0.107$, $d = 0.94$) No other differences were observed in the inactive and active group.

Table 5.4 Daily macronutrient intake

		Active			Inactive		
		Protein	Fat	CHO	Protein	Fat	CHO
Exp. day (%)	Ex.	14 ± 3	35 ± 12	51 ± 13	16 ± 9	35 ± 10	49 ± 8
	Con.	18 ± 5	38 ± 12	44 ± 12	15 ± 4	38 ± 12	47 ± 14
Day 1 (%)	Ex.	15 ± 4	33 ± 8	52 ± 10	12 ± 2	39 ± 20	49 ± 19
	Con.	14 ± 4	32 ± 5	54 ± 3	16 ± 5	32 ± 11	52 ± 14
Day 2 (%)	Ex.	16 ± 3	35 ± 11	49 ± 11	13 ± 4	31 ± 4	56 ± 8
	Con.	16 ± 6	37 ± 7	47 ± 8	15 ± 4	37 ± 7	48 ± 7
Day 3 (%)	Ex.	15 ± 3	28 ± 9	57 ± 8	13 ± 5	30 ± 6	57 ± 4
	Con.	19 ± 9	33 ± 10	48 ± 15	18 ± 5	33 ± 14	49 ± 14

N=8 for active and N=7 for inactive; values presented as mean ± SD. Exp. = experimental; Ex. = exercise condition; Con. = control condition; CHO = carbohydrates.

5.3.9 Percentage of energy compensation

Percentages of energy compensation are presented in figure 5.4 and their 95% confidence intervals in figure 5.5. There were no compensatory responses for the active group at the *ad libitum* lunch (13 ± 59%; 95% Cl -32 to 57%; Figure 5.5: panel A), the end of the experimental day (50 ± 265%; 95% Cl -151 to 251%; Figure 5.5: panel B) or subsequent day one (-53 ± 122%, 95% Cl -146 to 39%; Figure 5.5: panel C), day two (27 ± 396%; 95% Cl -273 to 372%; Figure 5.5: panel D) or day three (36 ± 258%; 95% Cl -159 to 232%; Figure 5.5: panel E). Similarly, there were no compensatory responses for the inactive group at the *ad libitum* lunch (47 ± 81%; 95% Cl -15 to 108%; Figure 5.5: panel A), the end of the experimental day (220 ± 427%; 95% Cl -104 to 544%; Figure 5.5: panel B) or day one (7 ± 338%, 95% Cl -250 to 263%; Figure 5.5: panel C), day two (137 ± 188%; 95% Cl -6 to 279%; Figure 5.5: panel D) or day three (34 ± 391%; 95% Cl -262 to 330%; Figure 5.5: panel E) after the experimental day. There were no group differences for the percentages of energy compensation for the *ad libitum* lunch $ip = 0.39$, $d = -0.51$), experimental day ($p = 0.40$, $d = -0.51$), day one ($p = 0.67$, $d = -0.25$), day two $ip = 0.54$, $d = -0.38$) or day three $ip = 0.99$, $d = 0.01$) after the experimental day. The cumulative percentage of energy compensation over the four days was also not different $ip = 0.48$, $d = -0.42$) between the active (61 ± 589%; 95% Cl -386 to 507%) and inactive (398 ± 1059%; 95% Cl -405 to 1200%) groups (Figure 5.5: panel F).

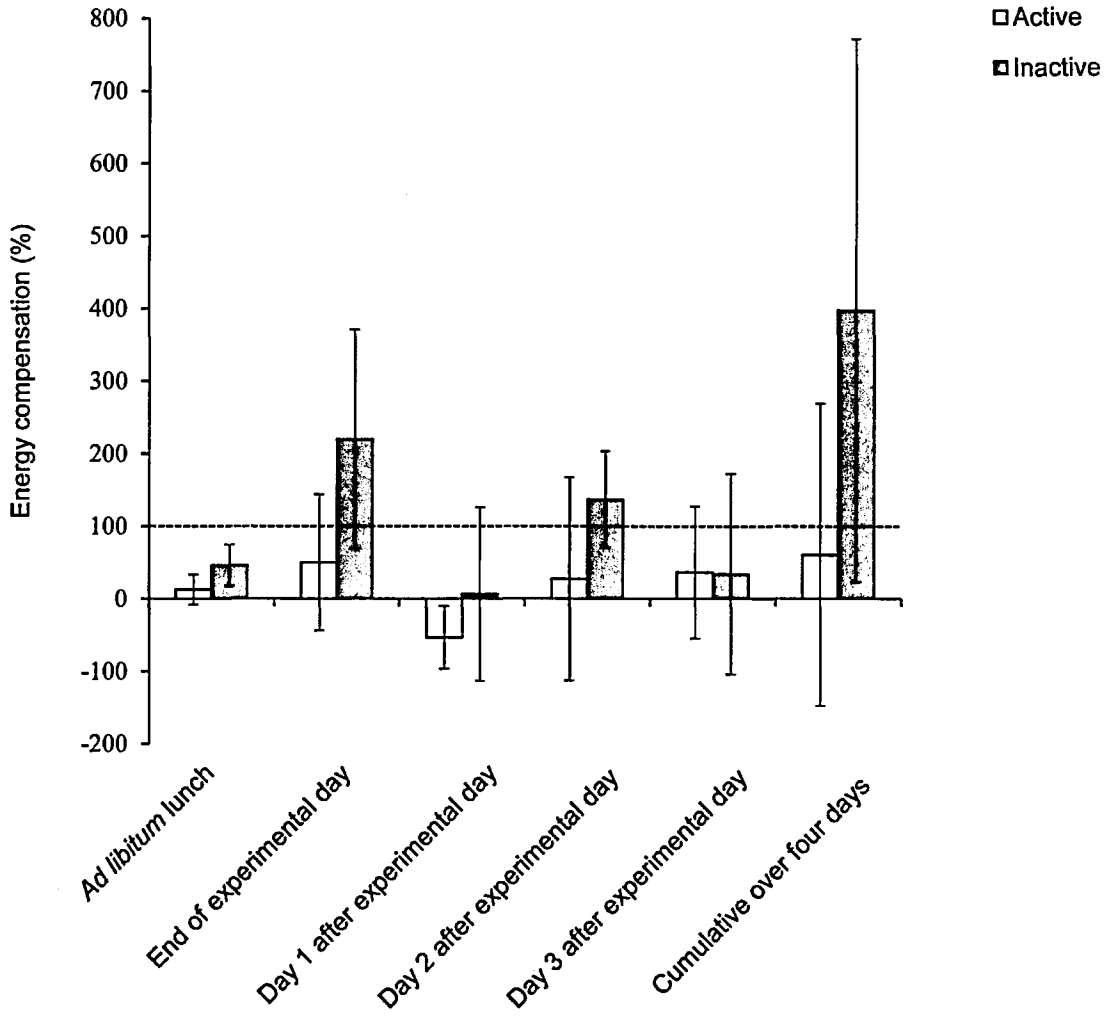


Figure 5.4 Percentages of energy compensation (N=8 for active and N=7 for inactive; means ± SEM); Exp. = Experimental. Dashed line indicates complete compensation (100%) of the exercise-induced energy expenditure.

if

5.3.10 Daily energy expenditure

One participant in the active group and one participant in the inactive group did not complete heart rate and accelerometer data collection for the full three days, therefore analyses are for 7 active and 7 inactive participants. There was only a group effect ($F(1,12) = 14.141$; $p = 0.003$, $d = 2.00$) for total free-living energy expenditure, indicating that active participants expended more energy than inactive over the course of the three days (mean difference = 3527 kJ; 95% Cl 2148 to 4906 kJ). There were no other main effects or interactions ($p > 0.05$) for free-living energy expenditure. Likewise there was only a group effect ($F(1,12) = 19.336$; $p = 0.001$, $d = 2.24$) for physical activity energy expenditure, indicating that the active group was more active than the inactive group (5244 ± 1791 kJ vs. 2189 ± 879 kJ) over the 3 days after the experimental days (Figure 5.6)

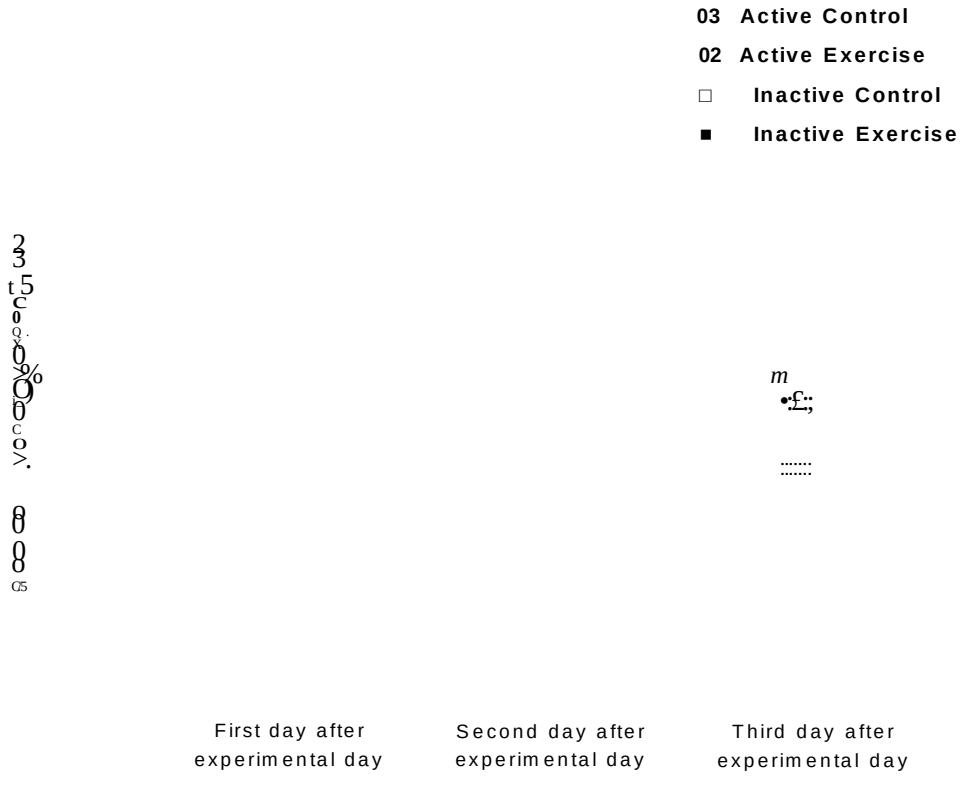


Figure 5.6 Daily physical activity energy expenditure (n=7 per group; means ± SEM).

5.4 Discussion - Women not using hormonal contraceptives (Study 2)

To the author's knowledge, the present study is the first to examine the effects of an acute bout of moderate-intensity aerobic exercise on immediate and subsequent three-day energy intake and expenditure in active and inactive women not using hormonal contraceptives. The main finding arising from this study is that there were no condition or group differences in energy intake and expenditure during the remainder of the experimental day or subsequent three days suggesting that there were no short-term compensatory responses to the exercise-induced energy deficit. However, during the experimental days there was a reduction in relative energy intake at lunch only in the active group.

As in the previous chapter, this study prescribed individual exercise intensities to reflect fitness status whilst aiming for both groups to cycle at approximately the same relative intensity. Despite no differences in the mean relative intensity of exercise in both groups, inactive participants tended to exercise at a higher relative intensity and perceive exercise as more strenuous. This can be explained by some inactive participants having a prescribed resistance equal to the lowest possible resistance in the cycle ergometer meaning that no downward adjustments could be made for these participants. Nevertheless, active participants expended more energy during exercise than inactive participants because they exercised at a higher absolute exercise intensity.

Participants' body mass did not change between conditions suggesting that participants remained in energy and fluid balance during the 4-week period between the first and the

second experimental day. To ensure participants remained in fluid balance water was provided *ad libitum*, and as anticipated, water consumption was higher during the exercise compared to the control experimental day. However, the participants' body mass at the start and end of the exercise period and the mean difference in water intake between conditions do not suggest water overconsumption and therefore it is unlikely that this difference had an impact on subsequent energy intake.

Consistent with previous research (Finlayson *et al.,* 2009; King *et al.,* 1996; Lluch *et al.,* 1998; Pomerleau *et al.,* 2004; Unick *et al.,* 2010), this study did not find any differences in subjective hunger ratings either between groups or conditions. It is possible that these findings are explained by the moderate exercise intensity (-50% of V_{O_2}max) used in this study because, as discussed in the previous chapter, male studies have provided strong evidence that the short-term suppression of hunger tends to occur only during and immediately after vigorous exercise (>60% of VCLmax) (Broom *et al.,* 2007, Broom *et al.,* 2009). Nevertheless, the relationship between exercise intensity and hunger has not been consistently reported in women. For instance, high-intensity (-70% of V02max) walking (Pomerleau *et al.,* 2004) and cycling (King *et al.,* 1996) did not have any effects on hunger after exercise whereas 30 minutes of intermittent treadmill exercise of 1 minute at high-intensity (70% of VC^max) alternating with 3 minutes of moderate-intensity (40% of V02max) (Reger *et al.,* 1984) and a combination of aerobic and muscle conditioning exercise (intensity not controlled) (Maraki *et al.,* 2005) has been shown to decrease and increase hunger, respectively. These studies suggest that exercise may not suppress hunger in the same way for men and women, however, findings from the present study and from the previous chapter do not support this and

suggest that 60 minutes of moderate-intensity cycling has no effect on hunger irrespectively of sex or habitual physical activity.

Energy intake during the *ad libitum* lunch meal did not differ between conditions or groups. This finding supports previous research that has consistently reported no difference in energy intake at the first meal after exercise in active (Finlayson *et al.*, 2009; Hagobian *et al.*, 2012; Lluch *et al.*, 1998; Lluch *et al.*, 2000; Larson-Meyer *et al.*, 2012) and inactive women (George & Morganstein, 2003; Maraki *et al.*, 2005; Reger *et al.*, 1984; Tsofliou *et al.*, 2003; Unick *et al.*, 2010). After adjusting for the energy expended during the exercise/rest period, the active group had a lower lunch REI after exercise than control, a finding observed in previous research with active women (Lluch *et al.*, 1998; Lluch *et al.*, 2000). In contrast with previous findings (Maraki *et al.*, 2005; Unick *et al.*, 2010), there was no difference in the inactive group lunch REI in the exercise and control condition but this discrepancy in results could be attributable to differences in participant characteristics and mode of exercise. This finding suggests that in the present study active participants had a higher exercise-induced energy deficit than inactive participants. This can be explained by the lower energy expended by the inactive group during the 60 min of exercise, however, the difference of the net exercise-induced energy expenditure between groups (320 kJ) does not account for all the difference between the exercise-induced energy deficit after lunch (623 kJ). The remaining difference can only be explained by changes in energy intake at the *ad libitum* lunch. Therefore, these results suggest that it is more difficult to attain and maintain a substantial exercise-induced energy deficit in inactive participants due to their lower energy expended during exercise and partial compensation during the *ad*

libitum lunch meal, which, in this study, accounted for approximately 47% of their net exercise-induced energy expenditure.

Total daily energy intake remained unaffected by exercise suggesting that contrary to men (chapter 4) there were no compensatory responses to an acute bout of moderate-intensity cycling in these women. These findings support previous studies that have reported no differences in energy intake after exercise and rest during the 24 h of the experimental day in active (Lluch *et al.*, 1998) and inactive women (Maraki *et al.*, 2005). One study has reported no effects of 30 and 60 min of continuous moderate-intensity (50% of $\dot{V}O_{2max}$) walking/jogging and 30 min of intermittent treadmill exercise with 1 min at high-intensity (70% of $\dot{V}O_{2max}$) alternating with 3 min at moderate-intensity (40% of $\dot{V}O_{2max}$) on energy intake over two days (Reger *et al.*, 1984). Additionally, Pomerleau *et al.* (2004) indicated that there were no effects of moderate-intensity (40% of $\dot{V}O_{2max}$) and high-intensity (70% of $\dot{V}O_{2max}$) walking on energy intake during the three days after exercise. Of note is that, Reger *et al.* (1984) study did not control for participants menstrual cycle, premenstrual symptoms and testing day of the week with experimental days being conducted between 5 to 9 days apart and Pomerleau *et al.*, (2004) only examined active women. Since energy intake in the present study was observed during the remainder of the experimental day and subsequent three days under more controlled conditions in active and inactive women, it provides evidence that, over this period of time, there were no exercise-induced delayed compensatory responses.

Similarly to energy intake, the calculated percentages of energy compensation for the different time periods in this study did not indicate any compensatory responses during or after the experimental day. Nevertheless, with the exception of day two after the experimental day (Figure 5.5: panel D), the 95% confidence intervals of the active

group are narrower than the ones of the inactive group a finding consistent with findings from chapter 4. This gives support to a possible tighter control of energy intake in active than inactive participants.

The inactive group consumed less energy from proteins over the four days of the exercise than the control condition that occurred with a trend for higher carbohydrate consumption during the exercise compared to the control condition. This could be explained by a motivation of inactive participants to seek specific foods to restore energy stores or preferences for tastes related with certain nutrients that would normally be associated with the carbohydrates needed to replenish the glycogen stores (Blundell *et al.*, 2003). The ability of an acute bout of exercise to improve psychological wellbeing (Deslandes *et al.*, 2009; Fox, 1999; Yeung, 1996) could also be related with changes in protein intake as lower energy intake of protein during the early follicular phase of the menstrual cycle (period over which the experimental studies were completed) has been associated with higher ratings of wellbeing in healthy women not taking oral contraceptives (Johnson *et al.*, 1995).

Total energy expenditure and physical activity energy expenditure during the free-living was greater in active than inactive participants, however, there were no differences between conditions suggesting that both groups maintained their physical activity. These results agree with findings from the previous chapter in men suggesting an acute bout of moderate-intensity cycling does not elicit compensatory changes in daily physical activity energy expenditure in premenopausal women not using hormonal contraceptives.

In summary, this study demonstrated that an acute bout of moderate-intensity aerobic exercise did not increase hunger or energy intake at the meal immediately after exercise, and induced a decrease in lunch relative energy intake after exercise only in the active group. Additionally, there were no exercise-induced compensatory responses in energy intake and expenditure during the remainder of the experimental day or subsequent three days. These findings provide information about the effects of exercise on the immediate and delayed energy intake and expenditure in active and inactive women not using hormonal contraceptives during the early to mid-follicular phase of the menstrual cycle and support the use of exercise in this population as a method to induce a short-term negative energy balance.

5.5 Results - Women taking oral contraceptives (Study 3)

5.5.1 Baseline characteristics

Participants' baseline characteristics are presented in table 5.5. Active participants had greater VCbmax and lower percentage of body fat than inactive participants *(p* < 0.05). There were no differences in age, stature, body mass, BMI and waist-to-hip ratio.

Table 5.5 Participants baseline characteristics

	Active	**Inactive**
Age (years)	22.6 ±3.6	22.3 ±3.2
Stature (m)	1.68 ±0.07	1.67 ±0.07
Body mass (kg)	61.4 ±4.4	60.1 ±4.3
BMI (kg-in'2)	21.9 ±1.3	21.6 ±2.0
Waist-to-hip ratio	0.73 ± 0.04	0.75 ± 0.04
Body fat (%) *	22.5 ±3.7	26.7 ±3.6
V 02l,,ax **(ml kg'1 min'1) ****	36.8 ±3.1	29.9 ±4.1

N=10 per group; values presented as mean ± SD.

BMI = body mass index; V02max = maximal oxygen consumption.

* Means significantly different *(p* < 0.05).

** Means significantly different *(p* < 0.01).

5.5.2 Exercise responses and energy expenditure on the experimental days

Relative intensity of exercise and ratings of perceived exertion were not different between active and inactive participants during the exercise (51.2 ± 2.2% vs. 54.0 ±

7.5% ofV02max;p = 0.27; RPE: 11.9 ± 1.6 vs. 11.7 ± 1.2;p = 0.79). There were no main or interaction effects for RER (p > 0.05) and only a condition (F(l,18) = 709.5; p < 0.001) effect for heart rate that, as anticipated, was different between the control and the exercise experimental day (72 ± 11 bpm vs. 131 ± 14 bpm, p < 0.001). Similarly, there was only a condition (p < 0.001) effect for the energy expenditure during the 60 minutes of exercise (1345 ± 195 kJ) and equivalent resting period (325 ± 41 kJ) and no differences between the net exercise-induced energy expenditure of active and inactive participants (1078 ± 132 kJ vs. 964 ± 239 kJ; p = 0.227, d = 0.60). There was a group effect for total energy expenditure (active vs. inactive: 6389 ± 1036 kJ vs. 4949 ± 841 kJ, p = 0.001, d = 1.61) and physical activity energy expenditure (active vs. inactive: 2780 ± 857 kJ vs. 1571 ± 727 kJ, p < 0.001, d - 1.60) during the remainder of the experimental days. There were no other main or interaction effects (p > 0.05).

5.5.3 Body mass and water consumption during the experimental days

Body mass and water consumption on the experimental days are presented in table 5.6. There were no main or interaction effects (p > 0.05) for body mass on the experimental days. There was a condition (p = 0.008, d = 0.51) effect for water consumption that was higher during the exercise (524 ± 332 mL) than the control condition (380 ± 257 mL). However, post hoc analyses did not show any differences between conditions for active participants (p = 0.18, d = 0.30) and only showed a higher water intake on the exercise than the control experimental day for inactive participants (p = 0.026, d = 0.67). There were no other main or interaction effects (p > 0.05).

Table 5.6 Body mass and water consumption on the experimental days

	Active		Inactive	
	Exercise	Control	Exercise	Control
Body mass (kg) - start	61.1 ± 5.6	61.1 ± 5.5	61.1 ± 4.3	60.6 ± 4.2
Body mass (kg) - end	61.0 ± 5.6	61.0 ± 5.5	61.0 ± 4.4	60.6 ± 4.2
Water consumption (mL)	538 ± 325	452 ± 271	510 ± 374	308 ± 248

N=10 per group; values presented as mean ± SD.

5.5.4 Environmental temperature and humidity

There were no condition (exercise vs. control: 21.2 ± 2.6 °C vs. 22.0 ± 3.5 °C, $p = 0.31$), group (active vs. inactive: 21.5 ± 2.7 °C vs. 21.7 ± 3.4 °C, $p = 0.86$) or interaction effects ($p = 0.35$) for temperature. Likewise there were no condition (exercise vs. control: 33.8 ± 9.5% vs. 33.1 ± 8.1%, $p = 0.65$), group (active vs. inactive: 34.3 ± 7.5% vs. 32.3 ± 9.4%, $p = 0.46$) or interaction effects ($p = 0.11$) for humidity.

5.5.5 Subjective ratings of hunger

There was a main effect of time ($p < 0.001$) for hunger ratings but there were no interactions or other main effects ($p > 0.05$) (Figure 5.7). Differences in hunger ratings were also evaluated using AUC values for the time before and after breakfast (08:45-09:00), the following hours until lunch (09:00-12:00), and the time before and after lunch (12:00-12:20). There was a main effect of time ($p < 0.001$) for hunger AUC values but no interactions or other main effects ($p > 0.05$).

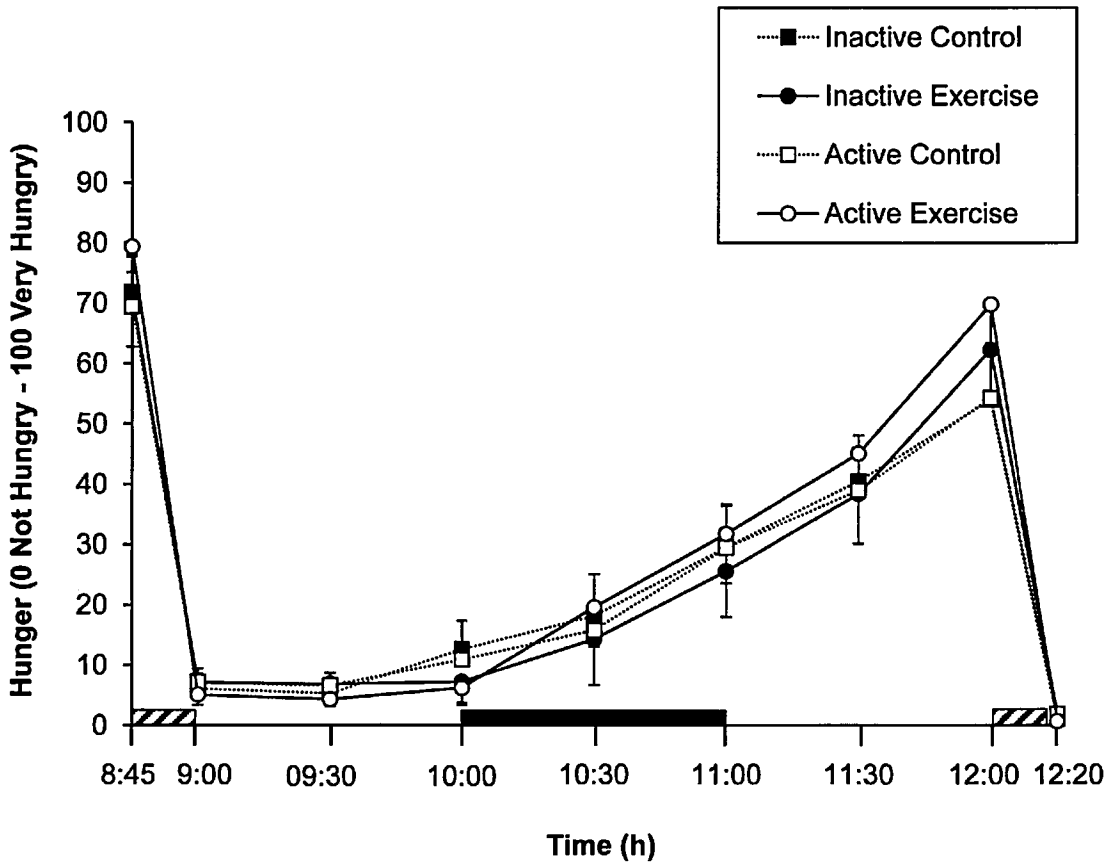


Figure 5.7 Subjective feelings of hunger (n=10 per group; means ± SEM). Hatched rectangles are consumption of meals; dark rectangle is equivalent to the 60 minutes cycling period.

5.5.6 *Ad libitum* lunch energy intake

The energy intake at the *ad libitum* lunch meal for active and inactive participants on both experimental days is presented in table 5.7. There was only a condition (p = 0.033, d = -1.30) effect for absolute energy intake at the *ad libitum* lunch (exercise vs. control: 3363 ± 668 kJ vs. 3035 ± 752 kJ), however, post hoc tests did not show any difference for the active (mean difference = 437 kJ; 95% CI -17 to 890 kJ, p = 0.069, d = 0.54) and inactive (mean difference = 222 kJ; 95% CI -187 to 631 kJ, p = 0.275, d = 0.42) groups. After adjustment of absolute energy intake for the energy expended during the 60 min of exercise/rest (relative energy intake, REI), there was a condition effect (F(1,18) =

19.723; $p < 0.001$, $d = -1.00$) with a lower REI in the exercise than the control condition (2019 ± 746 kJ vs. 2710 ± 712 kJ). Post hoc analysis demonstrated a difference in relative energy intake between conditions in the active (mean difference = -641 kJ; 95% CI -1124 to -158 kJ, $p = 0.019$, $d = -0.76$) and inactive (mean difference = -742 kJ; 95% CI -1203 to 280 kJ, $p = 0.007$, $d = -1.39$) groups.

Table 5.7 *Ad libitum* lunch meal energy intake

	Active	Inactive
Absolute EI Exercise condition (kJ)	3621 ± 853	3108 ± 342
Absolute EI Control condition (kJ)	3184 ± 841	2886 ± 706
Relative EI Exercise condition (kJ)	2234 ± 938*	1804 ± 379**
Relative EI Control condition (kJ)	2875 ± 828	2546 ± 701

N=10 per group; values presented as mean ± SD; EI = energy intake.

* Means significantly different from control condition ($p = 0.019$, $d = -0.76$).

** Means significantly different from control condition ($p = 0.007$, $d = -1.39$).

5.5.7 Daily energy intake

Daily energy intake for both groups is shown in figure 5.8. One participant in the inactive group did not complete the full four-day food diary, therefore analyses were made with 10 active and 9 inactive participants. There was a time ($p = 0.003$) and group ($p = 0.036$) effect and a trend for a condition x group x time interaction ($p = 0.056$) for daily energy intake. Pairwise comparisons showed that energy intake was greater on the experimental days (10180 ± 1670 kJ) compared to the subsequent first (8535 ± 2511 kJ,

p = 0.027, d = 0.81), second (8531 ± 2330 kJ, p = 0.022, d - 0.84) and third (8364 ± 2459 kJ, p = 0.024, d = 0.91) days and that inactive participants had a higher mean energy intake over the four days than the active group (9431 ± 1168 kJ vs. 8385 ± 1364 kJ, p = 0.036, d = -0.86). Post hoc analysis did not show any differences in the active group and inactive participants had only a decrease in energy intake on the first day after the exercise experimentalday compared withthe same day of the control condition (mean difference = -1974 kJ;95% Cl -1048 to-2900 kJ,p = 0.002, <7= -0.89).

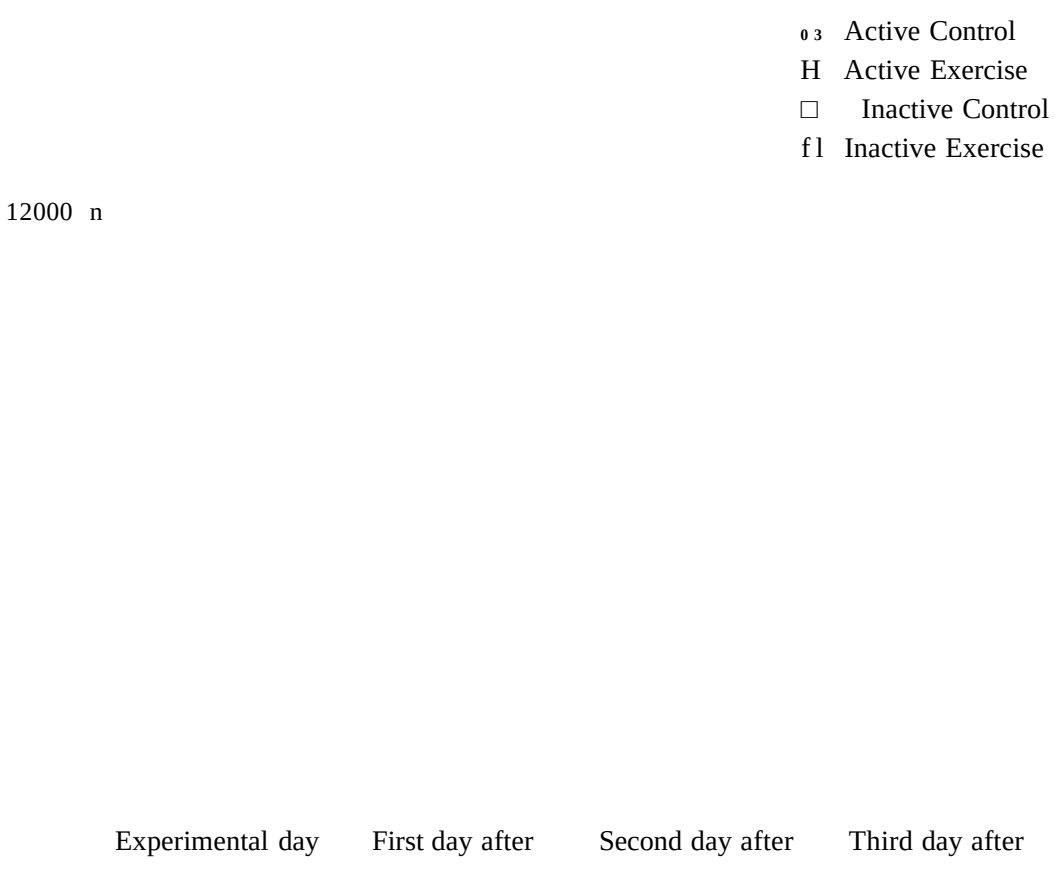


Figure 5.8 Daily energy intake (n=10 for active and n=9 for inactive; means ± SEM). *Means significantly different between conditions (p < 0.05);

5.5.8 Daily macronutrient intake

Daily macronutrient intake for both groups is shown in table 5.8. There were no main or interaction effects for the percentage of energy consumed from protein, fat and carbohydrate ($p > 0.05$) but there was a trend for condition x group interaction for energy consumed from fat ($p = 0.055$, $d = -1.35$). Post hoc analysis demonstrated that only the active group consumed less fat in the exercise than the control condition (mean difference = -6%; 95% CI -11 to -2%, $p = 0.012$, $d = -1.10$). No other differences were observed in the inactive and active group.

Table 5.8 Daily macronutrient intake

		Active			Inactive		
		Protein	Fat	CHO	Protein	Fat	CHO
Exp. day (%)	Ex.	19 ± 10	32 ± 12	49 ± 15	19 ± 6	32 ± 9	49 ± 12
	Con.	14 ± 4	34 ± 13	52 ± 11	16 ± 4	36 ± 8	48 ± 9
Day 1 (%)	Ex.	16 ± 5	33 ± 14	51 ± 12	16 ± 5	36 ± 8	48 ± 10
	Con.	14 ± 5	35 ± 11	51 ± 12	17 ± 5	35 ± 10	48 ± 11
Day 2 (%)	Ex.	18 ± 7	32 ± 8	50 ± 8	16 ± 6	34 ± 12	50 ± 7
	Con.	15 ± 4	34 ± 8	51 ± 8	16 ± 3	31 ± 8	53 ± 10
Day 3 (%)	Ex.	18 ± 5	28 ± 7	54 ± 7	15 ± 6	34 ± 12	51 ± 9
	Con.	16 ± 5	38 ± 5	46 ± 8	15 ± 5	32 ± 6	53 ± 9

N=10 for active and N=9 for inactive; values presented as mean ± SD. Exp. = experimental; Ex. = exercise condition; Con. = control condition; CHO = carbohydrates.

5.5.9 Percentage of energy compensation

Percentages of energy compensation are presented in figure 5.9 and their 95% confidence intervals in figure 5.10. There were no compensatory responses for the active group at the *ad libitum* lunch (43 ± 67%; 95% CI - 3 to 88%; Figure 5.10: panel A), the end of the experimental day (109 ± 208%; 95% CI - 32 to 250%; Figure 5.10: panel B) or subsequent day one (53 ± 346%, 95% CI - 182 to 287%; Figure 5.10: panel C), day two (-91 ± 293%; 95% CI - 289 to 107%; Figure 5.10: panel D) or day three (34 ± 267%; 95% CI - 147 to 215%; Figure 5.10: panel E) after the experimental day. Conversely, there was a negative compensation in the inactive group on the first day after the experimental day (-176 ± 150%, 95% CI - 278 to -74%; Figure 5.10: panel C) but no other compensatory responses at the *ad libitum* lunch (16 ± 67%; 95% CI - 30 to 61%; Figure 5.10: panel A), the end of the experimental day (-49 ± 216%; 95% CI - 196 to 98%; Figure 5.10: panel B) or subsequent day two (161 ± 371%; 95% CI -91 to 413%; Figure 5.10: panel D) or day three (1 ± 253%; 95% CI -171 to 173%; Figure 5.10: panel E) after the experimental day. There were no group differences for the percentages of energy compensation for the *ad libitum* lunch ($p = 0.63$, $d = 0.42$), experimental day ($p = 0.08$, $d = 0.78$), day one ($p = 0.053$, $d = 0.90$), day two ($p = 0.21$, $d = -0.80$) or day three ($p = 0.40$, $d = 0.13$) after the experimental day. The cumulative percentage of energy compensation over the four days was also not different ($p = 0.32$, $d = 0.26$) between the active (104 ± 718%; 95% CI -383 to 592%) and inactive (-62 ± 631%; 95% CI -494 to 366%) groups (Figure 5.10: panel F).

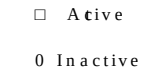


Figure 5.9 Percentages of energy compensation (N=10 for active and N=9 for inactive; means ± SEM); Exp. = Experimental. Dashed line indicates complete compensation (100%) of the exercise-induced energy expenditure.

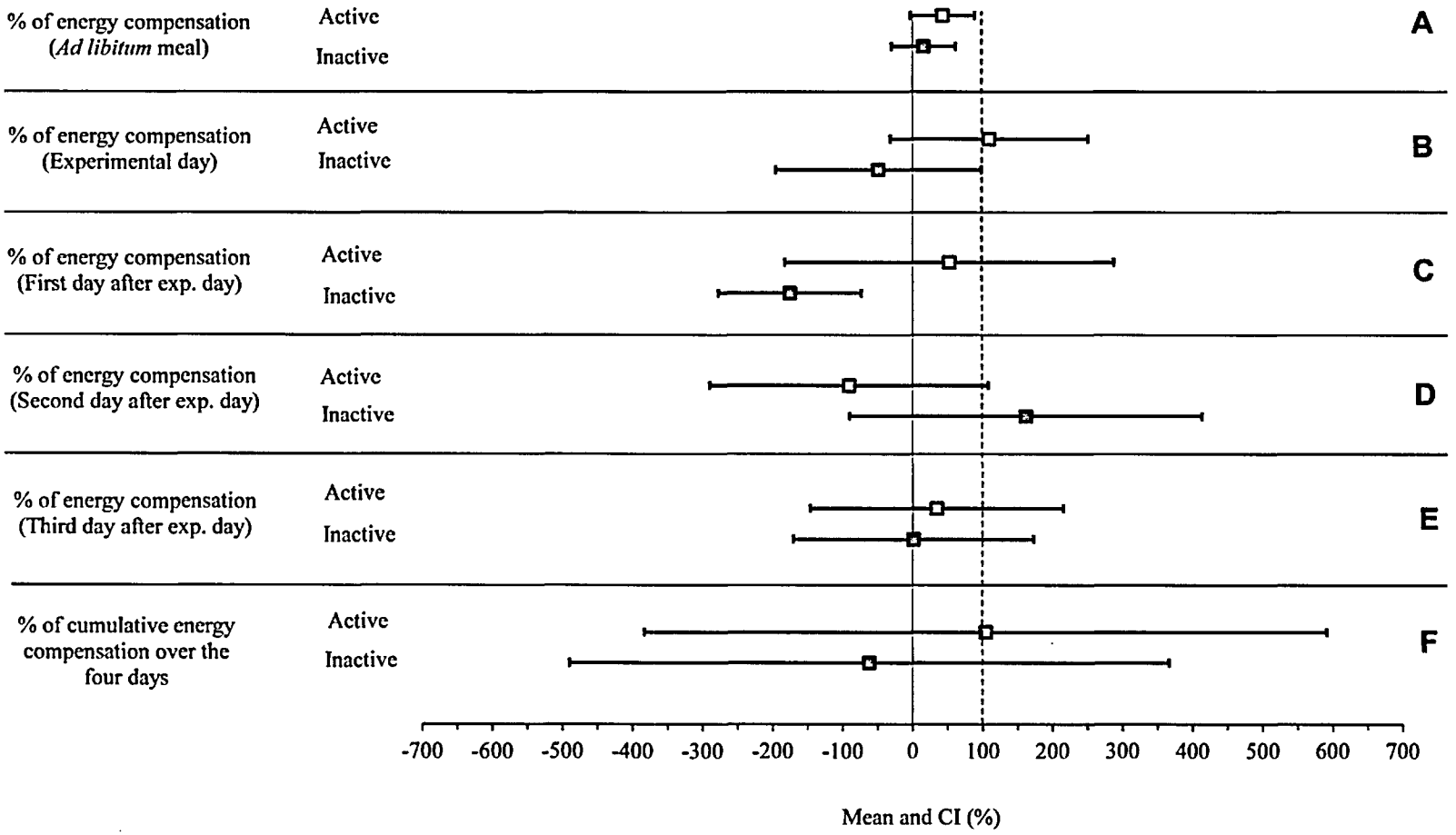


Figure 5.10 Mean percentages of energy compensation and 95% confidence intervals (N=10 for active and N=9 for inactive). Dashed line indicates complete compensation (100%) of the exercise-induced energy expenditure.

5.5.10 Daily energy expenditure

There was only a group effect ($F(1,18) = 15.817$; $p = 0.001$, $d = 1.63$) for total free-living energy expenditure, indicating that active participants expended more energy than inactive over the course of the three days (mean difference = 1573 kJ; 95% CI 597 to 2548 kJ). There were no other main effects or interactions ($p > 0.05$) for free-living energy expenditure. Likewise there was only a group effect ($F(1,18) = 21.763$; $p < 0.001$, $d = 1.73$) for physical activity energy expenditure, indicating that the active group was more active than the inactive group (3639 ± 787 kJ vs. 2363 ± 767 kJ) over the 3 days after the experimental days (Figure 5.11).

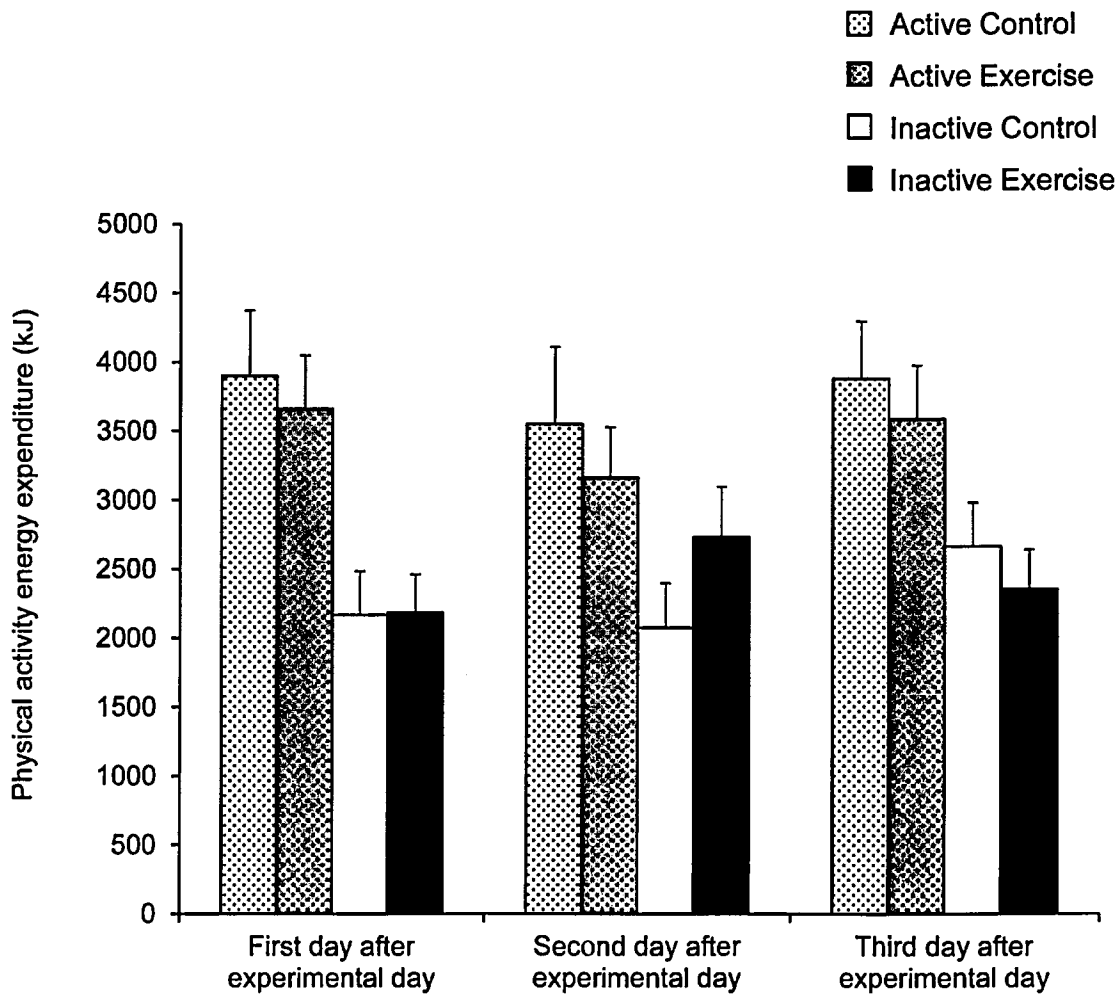


Figure 5.11 Daily physical activity energy expenditure (n=10 per group; means ± SEM).

5.6 Discussion - Women taking oral contraceptives (Study 3)

To the author's knowledge, the present study is the first to examine the effects of an acute bout of moderate-intensity aerobic exercise on immediate and subsequent three-day energy intake and expenditure in active and inactive women taking oral contraceptives. The main finding arising from this study is that exercise did not elicit changes in energy intake over the four days in active participants and induced a suppression of energy intake on the first day after the experimental day in inactive participants. Additionally, no differences were observed in both groups' physical activity energy expenditure between conditions suggesting that there were no acute compensatory changes to physical activity.

In contrast with the previous studies presented within this thesis, there were no differences between the net exercise-induced energy expenditure in active and inactive participants in the present study. This occurred despite both groups exercising at the same relative intensity and is possibly explained by the differences between groups' aerobic capacity being smaller than expected. There were no changes in body mass between conditions suggesting that similar to previous studies participants remained in energy and fluid balance during the period of time between the first and the second experimental day. Moreover, participants remained in fluid balance during the laboratory period of the experimental days as no changes were observed in body mass from the start to the end of the exercise/rest periods.

There were no differences in subjective hunger ratings either between groups or conditions in this study. This finding is in agreement with studies one and two of this thesis and recent studies with active (Finlayson *et al.*, 2009) and inactive women (Unick

et al., 2010). As previously discussed, the relationship between exercise intensity and hunger has not been consistently reported in women therefore it is difficult to ascertain if this finding is attributable to the moderate exercise intensity (≈50% of $\dot{V}O_{2max}$) used in this study. This is reinforced by the different outcomes of studies using cycling (King *et al.*, 1996), running (Reger *et al.*, 1984) and a combination of aerobic and resistance exercise (Maraki *et al.*, 2005) suggesting that, in women, the acute effect of exercise on hunger may also be determined by the type of exercise undertaken.

There was an overall condition effect on energy intake at the *ad libitum* lunch meal that was greater during the exercise than the control experimental day. However, this difference was not visible when differences were calculated for each group with only a trend being demonstrated in the active group. This may be explained by exercise-induced changes in the hedonic response to food, a finding reported in a previous study suggesting that active lean women who are prone to increase their energy intake have an increased implicit wanting for, and perceived palatability of, food (Finlayson *et al.*, 2009). Nevertheless, adjustment of energy intake for the energy expended during the exercise/rest period showed that both groups had a lower REI after exercise than control, suggesting that, similar to previous research in active (Pomerleau *et al.*, 2004) and inactive women (Unick *et al.*, 2010), participants maintained a short-term negative energy balance.

In this study, there were no differences in energy intake during the remainder of the experimental day or subsequent three days in the active group a finding consistent with the only study (to the author's knowledge) examining the effects of exercise on subsequent three days energy intake (Pomerleau *et al.*, 2004). Conversely, the inactive

group had a lower energy intake on the first day after the exercise experimental day compared with control and no other differences in the remaining days. This is a novel finding and is consistent with the study in men (chapter 4) as despite its large associated confidence intervals, the mean percentages of energy compensation suggest a suppression of energy intake in the first two days after exercise before the compensation on the third day. In the present study the mean percentages of energy compensation for the experimental day and subsequent two days have a similar pattern to study 1 as the values suggest that the active group entirely compensated on the experimental day and the inactive participants had a substantial positive compensation on the second day after the experimental day. These suggestions were, however, not confirmed with statistical significance so it is possible that this study was not able to detect these differences in energy intake due to lack of statistical power.

There were no differences between daily macronutrient intake in the exercise and control condition in the inactive group. However, the active group consumed less energy from fat over the four days of the exercise than the control condition, which is possibly explained by being more motivated to eat foods associated with restoring the expended energy (Blundell *et al.*, 2003). Total energy expenditure and physical activity energy expenditure during the free-living period of the study were not different between conditions suggesting that both groups maintained their physical activity. These results agree with findings from the previous studies suggesting that as in men, and women not using hormonal contraceptives an acute bout of moderate-intensity aerobic exercise does not elicit compensatory changes in daily physical activity energy expenditure in premenopausal women taking oral contraceptives.

In summary, this study demonstrated that an acute bout of moderate-intensity aerobic exercise did not increase hunger or energy intake at the meal immediately after exercise, and induced a decrease in lunch relative energy intake after exercise in both groups. There were no differences in active participants' daily energy intake over the four days whereas the inactive group decreased their daily energy intake on the first day after the exercise experimental day compared to control suggesting a delayed exercise-induced suppression of energy intake. Moreover, there were no compensatory changes in daily physical activity energy expenditure. These findings provide information about the effects of exercise on the immediate and delayed energy intake and expenditure in active and inactive women taking oral contraceptives and support the use of moderate-intensity aerobic exercise as a method to induce a short-term negative energy balance, particularly in inactive women.

Chapter 6: Effects of a 12-week aerobic exercise intervention on 7-day energy intake and energy expenditure in inactive men (study 4)

6.1 Introduction

Exercise is an important strategy to reduce or maintain body mass due to its potential to create a negative energy balance while improving several health-related outcomes (Jakicic & Otto, 2006). However, its role is often diminished by the perception that exercise does not provide a significant reduction in body mass (Donnelly & Smith, 2005) or, as discussed by Blundell (2009), by misleading and discouraging public health messages from the media such as "Health Warning: exercise makes you fat". The latter was partly based on the misinterpretation of previous research that suggested that compensatory responses may be responsible for the high inter-individual variability observed in exercise-induced reductions in body mass and fat tissue (Caudwell *et al.,* 2009; King *et al.,* 2008). In addition, participants' adherence to exercise interventions is often suggested as an explanation for this variability (Byrne *et al.,* 2006; Colley *et al.,* 2008; Wing, 1999) but even when this is accounted for, differences in body mass and fat loss are still observed (Caudwell *et al.,* 2013b; King *et al.,* 2009; King *et al.,* 2008). Compensatory changes in energy intake and non-exercise energy-expenditure have also been suggested to explain this variability (Blundell *et al.,* 2003; King *et al.,* 2007), however, the extent to which they contribute to individual differences in body mass or fat loss is unclear.

Most studies that have examined the effect of chronic exercise on energy intake for one to two weeks support that men can partially compensate for the exercise-induced energy expenditure by increasing energy intake (Staten, 1991; Stubbs *et al.*, 2004b; Whybrow *et al.*, 2008). Based on these findings, it could be hypothesised that a possible increase in energy intake towards a full or at least higher percentage of energy compensation would occur during subsequent weeks, however, studies examining this relationship with exercise interventions lasting from 6 to 12 weeks did not observe any compensatory increases in energy intake (Broeder *et al.*, 1992; Caudwell *et al.*, 2013a; Caudwell *et al.*, 2013b; Martins *et al.*, 2007b). This tendency for a lack of an effect of chronic exercise on energy intake has also been consistently reported in long-term (> 4 months) exercise intervention studies (Donnelly *et al.*, 2003; Frey-Hewitt *et al.*, 1990; Van Etten *et al.*, 1997; Westerterp *et al.*, 1992). Nevertheless, it is important to consider that some studies did not provide clear definitions of participants' activity status (Broeder *et al.*, 1992; Caudwell et al., 2013b; Staten, 1991; Stubbs *et al.*, 2002a; Stubbs *et al.*, 2004b; Van Etten *et al.*, 1997; Whybrow *et al.*, 2008), did not supervise exercise sessions (Martins *et al.*, 2007b), assessed energy intake through a food-frequency questionnaire (Drenowatz *et al.* 2012), and were not specifically designed to investigate the effects of chronic exercise on energy intake (Broeder *et al.*, 1992; Leon *et al.*, 1979), limiting the interpretation of findings.

In contrast, studies examining the effects of exercise intervention on non-exercise physical activity are equivocal. Some studies have reported exercise to induce decreases in non-exercise energy expenditure (Colley *et al.*, 2010; Goran & Poehlman, 1992; Manthou *et al.*, 2010; Meijer *et al.*, 1999; Meijer *et al.*, 2000; Morio *et al.*, 1998; Wang & Nicklas, 2011), whereas others reported no changes (Church *et al.*, 2009; Hollowell

et al., 2009; Keytel *et al.*, 2001; Turner *et al.*, 2010; Van Etten *et al.*, 1997), an increase (Hunter *et al.*, 2000; Racette *et al.*, 1995) and even mixed findings between groups (Manthou *et al.*, 2010; Meijer *et al.*, 1991; Rosenkilde *et al.*, 2012). Differences may be explained by participants characteristics (e.g. age), exercise characteristics (e.g. intensity), and different measurement methods (e.g. accelerometers vs. physical activity diaries). Regarding the latter, several of the above studies have assessed physical activity through physical activity diaries (McLaughlin *et al.*, 2006; Manthou *et al.*, 2010; Morio *et al.*, 1998; Racette *et al.*, 1995) which have questionable validity and reliability in the assessment of energy expenditure (Andre & Wolf, 2007).

There is a clear need to investigate the chronic effects of exercise on energy intake and physical activity energy expenditure with improved measurement methods. To overcome some of the limitations of previous research, this study examined the effects of a supervised moderate-intensity 12-week exercise intervention on 7-day energy intake and expenditure using weighed food diaries and Actihearts, respectively. Additionally, this study only examined inactive men because physical inactivity is a major risk factor for the development of diseases such as obesity and increased mortality from all causes (Haskell *et al.*, 2009), and the difficulties of controlling the variables such as menstrual cycle, premenstrual symptoms and use of hormonal contraceptives.

6.2 Methods

6.2.1 Participants

This study was advertised as a 12 week exercise intervention to help people become more active and examine the relationship between exercise, genetics and mood. This was the case because participants were also taking part in a separate genotyping study which was not part of this PhD thesis. As an incentive the advert mentioned that after successful completion of the study participants would receive a 3 months free gym membership. Seventy-eight participants requested more information about the study from which sixty-nine maintained interest. Twelve participants met the inclusion criteria for the study but one participant withdrew from the study because he was not willing to follow protocol, therefore, only a total of 11 inactive men completed the study (Figure 6.1).

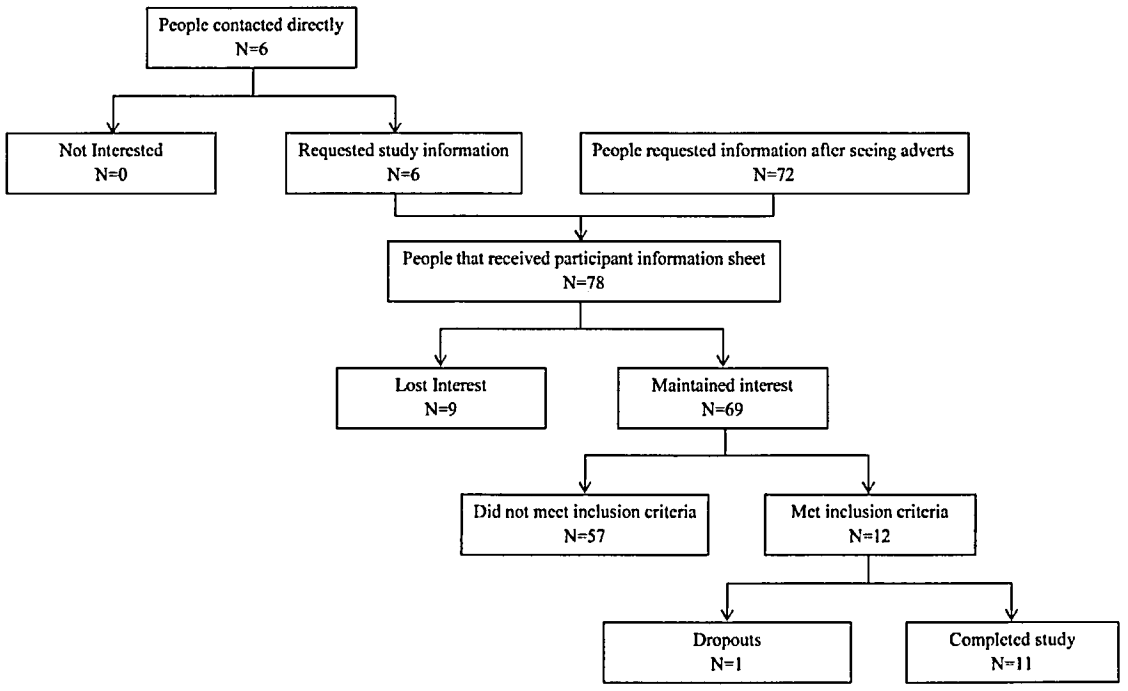


Figure 6.1 Consort diagram of the study.

Participants were recruited according to the following inclusion criteria:

- Male aged 18-40 years;
- Normal, Overweight or Obese (body mass index between 18.5 and 35 kg/m2);
- Healthy (no known chronic diseases);
- Non-smokers;
- Not dieting;
- Body mass stable (±2 kg) for 6 months prior to the study;
- Not taking medications that could affect food intake or metabolism.

As previously discussed in section 3.2, participants were excluded if deemed highly restrained eaters (scoring a raw score of more than 18 and a percentage above 66%). No participants had an individual score above these values and the mean group baseline score for cognitive restraint based on the revised version of the Three-Factor Eating Questionnaire (Karlsson *et al*., 2000) was 10.5 ± 2.1 (24.7 ± 11.5%). Self-reported weekly physical activity assessed by a modified version of Godin Leisure-Time Exercise Questionnaire (GLTEQ) (Godin & Shepard, 1985) was used to exclude participants who engaged in regular exercise and undertaking at least 150 minutes of moderate-intensity physical activity per week. Veracity of self-reported measures of physical activity was confirmed with baseline Actiheart data with all participants having a physical activity level within the sedentary to light activity lifestyle range (1.40-1.69) (WHO, 2004). The mean group physical activity level was 1.51 ± 0.1.

6.2.2 Study design

Participants who remained interested after reading the participant information sheet were contacted by telephone or e-mail for an initial screening, after which they were invited for an initial preliminary visit. During this visit participants were given a tour of the university facilities, explained in further detail the study requirements, any questions and doubts they had were answered, and an informed consent form (Appendix 3), health screen (Appendix 4), physical activity (Appendix 5), eating behaviour (Appendix 6) and food cravings (Appendix 12) questionnaires were completed. Participants who met the inclusion criteria were then invited for a second preliminary visit booked in the morning after a 10-hour overnight fast with only water consumption permitted. This visit involved the measurement of resting blood pressure and heart rate, collection of finger prick capillary blood samples, anthropometric measurements, completion of the Astrand-Rhyming cycle ergometer test, and an explanation of dietary recording and the use of the Actiheart. Participants then wore the Actiheart and recorded their food consumption for a week before starting the supervised 12 week exercise programme. After completion of the exercise intervention participants visited the lab for a post-intervention assessment conducted at the same time of day and under the same conditions as the second preliminary visit. This visit included the completion of the eating behaviour and food cravings questionnaires and the same measures undertaken during the second preliminary visit. The seven day assessment of post-exercise intervention free-living energy intake and expenditure was undertaken starting in the same day of the week as in the pre-intervention assessment. At the end of the study participants were debriefed about the true purpose of the study. A schematic representation is presented in figure 6.2.

Figure 6.2 Schematic representation of the study.

6.2.3 Eating behaviour and food cravings

Participants were asked to complete the revised version of the Three-Factor Eating Questionnaire (TFEQ-R18) (Karlsson *et al.*, 2000 - Appendix 6) at the preliminary and post-intervention visits to assess if there were any changes in three aspects of eating behaviour, namely their cognitive restraint (conscious restriction of food intake in order to control body mass), uncontrolled eating (tendency to eat more than usual due to a loss of control over intake accompanied by subjective feelings of hunger) and emotional eating (inability to resist emotional cues). Participants' food cravings over the previous month were also measured at the preliminary and post-intervention visits using the validated Food Craving Inventory (FCI) (White *et al.*, 2002). This is important because food cravings have also been implicated as an important factor influencing appetite control (Burton *et al.*, 2007; Gilhooly *et al.*, 2007; Martin *et al.*, 2008). The FCI is a 28-item self-report measure of general and specific food cravings scored on a 1-5 Likert scale (1 = Never, 5 = Almost every day). It consists of four subscales: carbohydrates/starches (8 items), fast-food fats (4 items), high-fat foods (8 items), and sweets (8 items) and defines food craving as an intense desire to consume a particular food (or food type) that is difficult to resist (White *et al.*, 2002). According to the same authors this definition adopts the perspective that food craving is an internal experience with cognitive and emotional (drive or motivational) properties. Additionally, the subscales of this inventory have shown that obese and non-obese participants differ across specific subscale scores but not in total scores (White *et al.*, 2002). For the present study, the FCI was modified slightly with substitution of typically American foods with English equivalents (e.g., replacing "hot dogs" with "sausage rolls") that contained similar macronutrient content. This ensured that the composition of each subscale remained unaffected. Total craving score was calculated as the mean of all

food items scores and the specific craving score as the mean score for the food items included in that subscale.

6.2.4 Resting heart rate and arterial blood pressure

Resting heart rate and arterial blood pressure were measured during the health screening by a calibrated oscillometric blood pressure monitor (Dash 2500, GE Healthcare, Finland) as mentioned in section 3.4.

6.2.5 Blood analyses

Finger prick capillary blood samples were collected after a 10 hour overnight fast and analysed with an enzymatic peroxidase dry chemistry method (Cholestech LDX System) to determine plasma total cholesterol (TC), high density lipoproteins (HDL), low density lipoproteins (LDL), non-high density lipoproteins (Non-HDL), triglycerides and glucose concentrations. The Cholestech LDX analyser has been previously validated (Carey *et al.*, 2006; Santee, 2002) with comparisons made against laboratory analysis showing correlations of 0.91 for TC, 0.77 for HDL-C, 0.88 for LDL-C and 0.98 for triglycerides (Parikh *et al.*, 2009). This method has a range of detection for TC (2.59-12.9 mmol/L), HDL-C (0.39-2.59 mmol/L), triglycerides (range 0.51-7.34 mmol/L), and glucose (range 2.78-27.8 mmol/L). LDL and Non-HDL are calculated from previous values as follows:

LDL = (TC) - (HDL) - (triglycerides ÷ 5)

Non-HDL = TC - HDL

6.2.6 Anthropometric measurements

Stature, body mass, waist and hip circumferences and body composition via a bioelectrical impedance analyser (InBody720, Derwent Healthcare Ltd, Newcastle upon Tyne, UK) were measured according to the procedures described in section 3.3

6.2.7 Astrand-Rhyming cycle ergometer test

As described in section 3.8.3, participant's maximum oxygen consumption was estimated using the Astrand-Rhyming submaximal cycle ergometer test (Astrand & Rhyming, 1954). This test was chosen because of its applicability to men of various ages who are not used to exercising and to reduce the overall risk involved in this study.

6.2.8 7-day free-living energy intake

Participants were instructed to weigh and record all items of food and drink consumed both at home and outside the home in food diaries, as described in section 3.14.

6.2.9 7-day free-living energy expenditure

Free-living energy expenditure for the seven days was estimated using an Actiheart (Cambridge Neurotechnology, Cambridge, UK) as described in section 3.15.

6.2.10 Exercise intervention

All supervised sessions were undertaken by the researcher at the Centre for Sports and Exercise Science (CSES) exercise suite based at Collegiate Hall. During the exercise

intervention, participants exercised for one hour a minimum of three times per week. Each session involved a 5-10 minute warm-up period, a 40 minute main exercise period at approximately 50-60% of heart rate reserve (HRR) and a 5-10 minute cool down period. Energy expended by each participant during the exercise sessions was estimated at week 1 and week 12 of the intervention using Keytel *et al.* (2005) heart rate prediction equation as follows:

$$EE\ (kJ/min) = -59.3954 + sex \times (-36.3781 + 0.271 \times age + 0.394 \times body\ mass + 0.404 \times \dot{V}O_{2max} + 0.634 \times HR) + (1 - sex) \times 0.274 \times age + 0.103 \times body\ mass + 0.380 \times \dot{V}O_{2max} + 0.450 \times HR)$$ [In this equation sex = 1 for men and 0 for women]

This equation improves on previous heart rate predictive equations (Hiilloskorpi *et al.*, 1999; Rennie *et al.*, 2001) by allowing adjustment for age, sex, body mass and fitness. In Keytel *et al.*, (2005) study this equation had a correlation coefficient between the measured and estimated energy expenditure of 0.913 and accounted for 83.3% of the variance in energy expenditure. To test the validity of this equation in a sample of inactive individuals, the author compared the exercise energy expenditure measured by indirect calorimetry for the 15 inactive participants of study 1 of this thesis with the predicted energy expenditure calculated with this equation. In this sample, the predicted energy expenditure underestimated the measured energy expenditure by a mean difference of -55.9 ± 406.1 kJ (-4.7 ± 23.6%) and had a Pearson correlation coefficient of 0.811.

It is important to note that each participant's exercise intensity and mode were also altered according to the participant's rating of perceived exertion and general feedback

(e.g. "Exercise is really easy but I feel a little bit of tightness in my calves"). Participants could choose to exercise on a treadmill, cycle ergometer, rower and elliptical ergometer. For the first three weeks participants completed three sessions per week, and thereafter these could increase to a maximum of four supervised sessions per week subject to the participant's choice. As the study progressed participants were encouraged to undertake unsupervised sessions to facilitate the transition from a supervised environment (during the study) to an unsupervised one (after completion of the study), however these were optional. These sessions were also undertaken when work or other commitments did not allow participants to attend the three sessions at the CSES exercise suit. For these sessions the researcher gave participants a heart rate monitor and a session plan for them to undertake in their own time. This required participants to input the day, time, type, duration and intensity (i.e. mean heart rate during each bout of exercise as displayed on the heart-rate monitor) of all exercise undertaken in the session plan sheet.

6.2.11 Statistical analyses

Sample size estimation was not undertaken because, to the author's knowledge, no published study has reported free-living energy intake and expenditure solely in inactive men after an exercise intervention of a similar duration. Regardless, as many participants were recruited and completed the study as possible within the time constraints of completing the PhD.

Data were analysed using the Statistical Package for the Social Sciences software for windows (SPSS 19.0, Chicago, IL, U.S.A.). Paired t-tests compared estimated exercise energy expenditure, mean exercise heart rate and rating of perceived exertion, body

composition, resting heart rate, arterial blood pressure, estimated maximum oxygen consumption, metabolic profile (total cholesterol, HDL, non-HDL, triglycerides, LDL, and fasting glucose), cognitive restraint, unrestrained eating, emotional eating, and food cravings before and after the exercise intervention. Two-way repeated measures ANOVA (Intervention x Time) compared energy intake, macronutrient intake and energy expenditure before and after the exercise intervention (Intervention effect) over the 7 days (Time effect). Cohen's *d* (standardised mean difference) effect sizes were calculated by dividing the difference between means by the pooled standard deviation thus reflecting differences expressed in standard deviation units. According to Cohen's (1988) guidelines, effect sizes may be conservatively interpreted as small (0.2), medium (0.5), and large (0.8) effects.

6.3 Results

6.3.1 Participants baseline characteristics

Participants' baseline characteristics are presented in table 6.1. Participant's age ranged from 19 to 34 years and body mass from 63.2 to 115.8 kg. Participants body mass index ranged from 19.7 to 33.8 kg-m" with participants being classified as lean (n=6), overweight (n=4) and obese (n=l).

Table 6.1 Participants baseline characteristics

	Baseline
Age (years)	25.5 ±4.8
Stature (m)	1.80 ±0.05
Body mass (kg)	79.9 ± 15.4
Body mass index (kg-m"2)	24.6 ±3.8
Body fat (kg)	14.6 ±8.5
Body fat (%)	17.4 ±7.3
Skeletal muscle mass (kg)	37.4 ±5.6
Skeletal muscle mass (%)	47.3 ±4.3
Waist circumference (cm)	82.9 ± 10.3
Hip circumference (cm)	99.6 ± 10.7
Visceral fat area (cm2)	71.9 ± 39.3
Estimated VC^max (ml/kg/min)	43.1 ±7.4

N=11; values presented as mean ± SD; V02max = maximum oxygen consumption.

6.3.2 Compliance and exercise energy expenditure

A diagram with information of the total number of exercise sessions that were completed as supervised, unsupervised or not undertaken is presented in figure 6.3. Individual compliance with the exercise sessions (supervised and unsupervised) was overall good with eight participants having 100% attendance, two participants 97% and one participant 81%. However, it is important to note that half of the unsupervised sessions were undertaken by only two participants that changed residences within the first two weeks of their exercise intervention. The new residences were an hour away from the CSES exercise suit by public transport making onsite exercise sessions logistically and financially challenging. This meant that contrary to most participants (n=9) that had sporadic unsupervised sessions these two participants had a very high percentage of unsupervised sessions (75% and 92%, respectively).

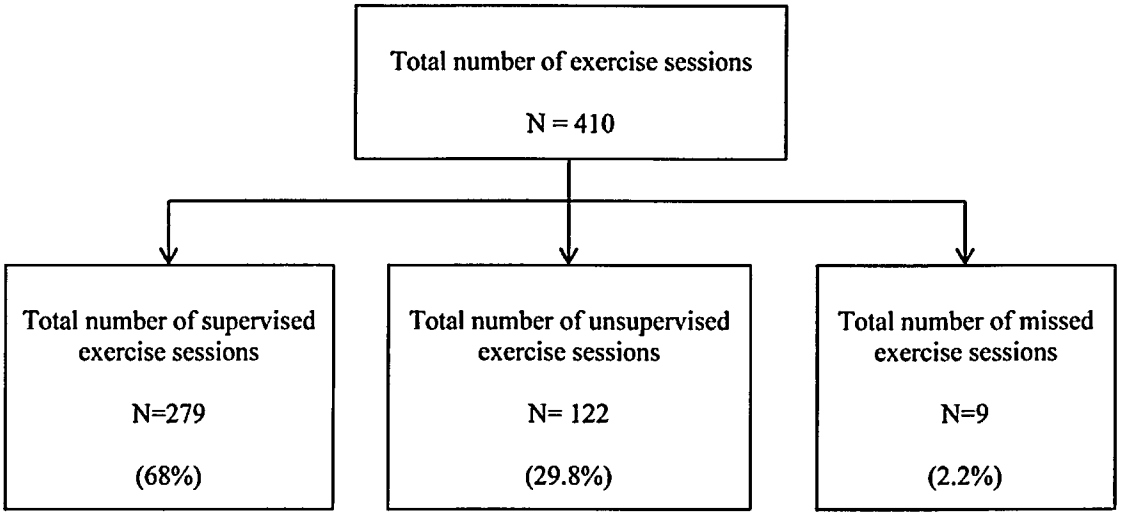


Figure 6.3 Diagram with total number and percentages of exercise sessions supervised, unsupervised and missed.

The mean heart rate, rating of perceived exertion and estimated energy expenditure during the exercise sessions in week 1 and week 12 are summarised in table 6.2. Mean exercise heart rate ($p = 0.031$, $d = 0.41$) and RPE ($p = 0.036$, $d = 0.29$) were higher in week 12 than in week 1 but there were no differences between the mean individual ($p = 0.646$, $d = 0.42$) or total weekly ($p = 0.370$, $d = 0.42$) exercise energy expenditure between the first and last week of the exercise intervention.

Table 6.2 Heart rate, ratings of perceived exertion and estimated energy expenditure of exercise sessions

	Week 1	**Week 12**
Mean heart rate of exercise sessions (bpm)*	144 ± 6	146 ± 6
Mean RPE of exercise sessions*	13.1 ± 1.0	13.4 ± 0.9
Mean energy expenditure of exercise sessions (kJ)	2477.8 ± 220.1	2526.6 ± 463.4
Total exercise energy expenditure of week (kJ)	7433.4 ± 660.3	7880.6 ± 2060.6

N=11 per group; values presented as mean ± SD. RPE= Rating of perceived exertion.

* Significant mean change ($p < 0.05$)

6.3.3 Anthropometric measurements

Mean group changes in the anthropometric measurements (from baseline to after the exercise intervention) are presented in figure 6.4. The 12-week exercise intervention induced reductions in body mass (mean difference = -1.6 ± 1.7 kg; $p = 0.011$, $d = -0.11$), BMI (mean difference = -0.4 ± 0.4 $kg \cdot m^{-2}$; $p = 0.008$, $d = -0.12$), waist circumference

(mean difference = -3.8 ± 2.8 cm; p = 0.001, d = -0.42), hip circumference (mean difference = -2.2 ± 1.7 cm; p = 0.002, d = -0.22), body fat (mean difference = -1.1 ± 1.4 kg; p = 0.025, d = -0.14) and percentage of body fat (mean difference = -1.1 ± 1.5%; p = 0.038, d = -0.16). No changes were observed in skeletal muscle mass (SMM) (mean difference = -0.4 ± 1.0%; p = 0.274, d = -0.07), percentage of SMM (mean difference = 0.5 ± 1.0%; p = 0.157, d = 0.12) or visceral fat area (mean difference = -1.1 ± 1.5%; p = 0.823, d = -0.01).

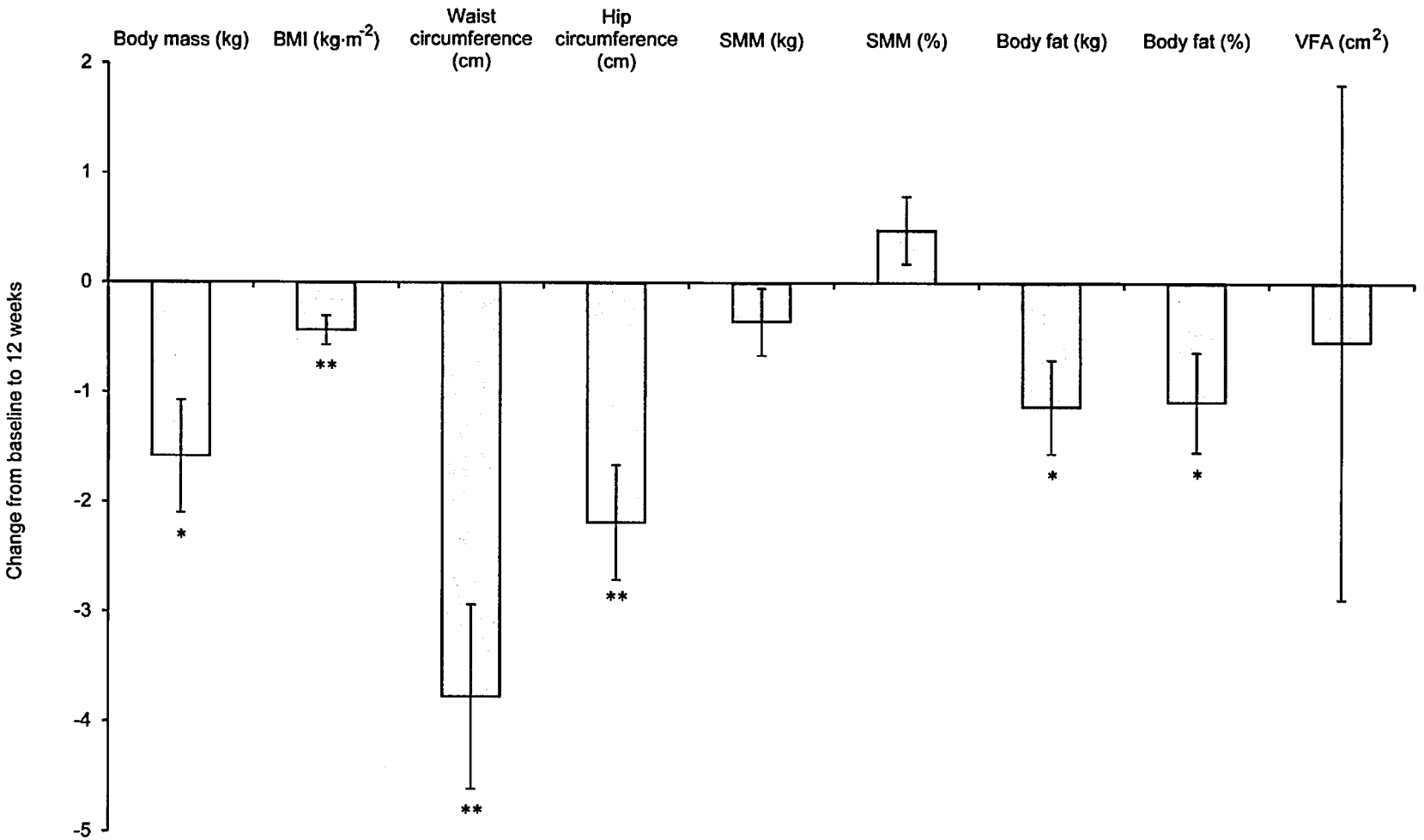


Figure 6.4 Mean group changes in the anthropometric measurements after 12 weeks (N=11; values presented as mean ± SEM); BMI= body mass index; SMM= skeletal muscle mass; VFA= visceral fat area. * Significant mean change ($p < 0.05$). ** Significant mean change ($p < 0.01$).

When assessing changes in body mass, body fat and SMM individually a large individual variation was observed (Figure 6.5). Individual changes from baseline to 12 weeks in body mass, body fat and SMM ranged from -5.2 to +1.2 kg, -3.3 to +1.9 kg and -2.4 to +1.0 kg, respectively.

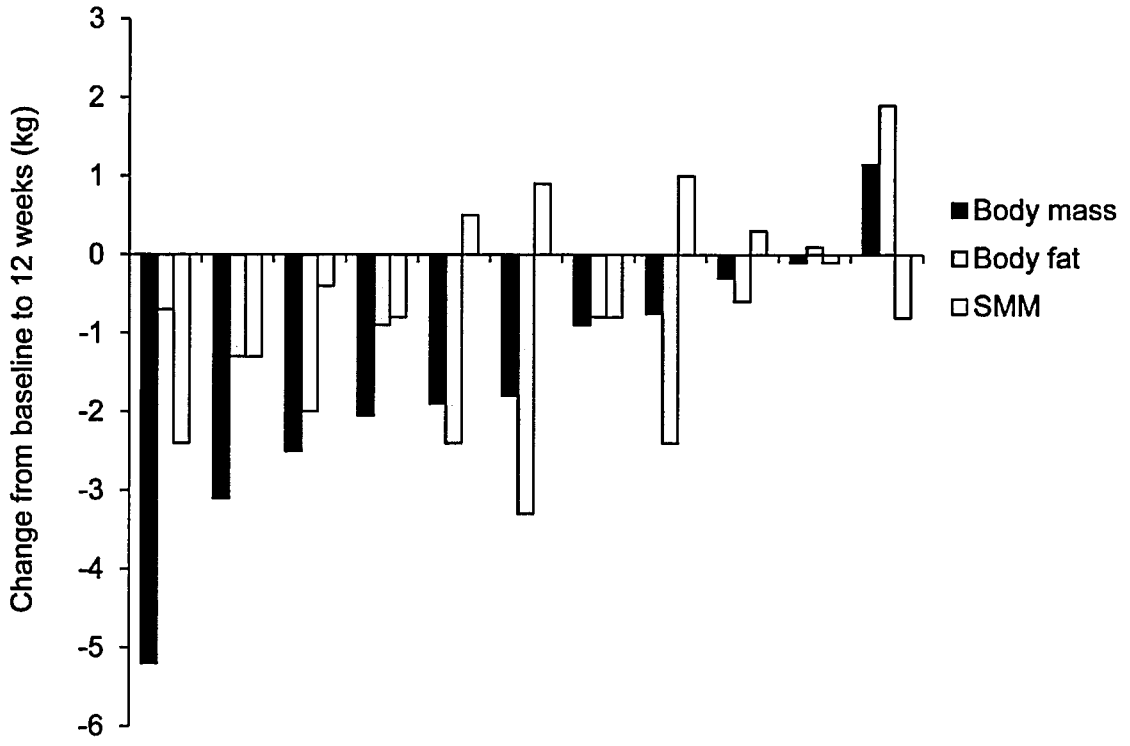


Figure 6.5 Individual changes in body mass, fat and skeletal muscle mass (SMM) in response to the 12 weeks exercise intervention. Each grouped three histograms represents values for one participant.

6.3.4 Resting heart rate, arterial blood pressure and cardiopulmonary fitness

Participants' resting heart rate, arterial blood pressure and estimated maximal oxygen consumption ($\dot{V}O_{2max}$) at baseline and after 12 weeks are presented in table 6.3. Resting diastolic blood pressure decreased (p = 0.021, d = -0.64) with the exercise intervention

whereas $\dot{V}O_{2max}$ increased (p = 0.001, d = 1.05). There were no differences between baseline and post-intervention values for resting heart rate (p = 0.657, d = -0.16) and systolic blood pressure (p = 0.186, d = -0.27).

Table 6.3 Resting heart rate, arterial blood pressure and maximal oxygen consumption.

	Baseline	**After intervention**
Resting heart rate (bpm)	58 ± 10	57 ± 8
Resting systolic blood pressure (mmHg)	120 ± 8	118 ± 8
Resting diastolic blood pressure* (mmHg)	70 ± 7	66 ± 6
Estimated $\dot{V}O_{2max}$ (ml/kg/min)**	43.1 ± 7.4	51.1 ± 8.4

N=11 per group; values presented as mean ± SD; $\dot{V}O_{2max}$ = maximal oxygen consumption; * Means significantly different ($p < 0.05$); ** Means significantly different ($p < 0.01$).

6.3.5 Metabolic profile

Participant's fasting plasma total cholesterol (TC), high density lipoproteins (HDL), low density lipoproteins (LDL), non-high density lipoproteins (Non-HDL), triglycerides and glucose concentrations before and after the 12 week exercise intervention are presented in table 6.4. Due to some participants having values outside the range of detection of the Cholestech LDX, sample size was different for TC and glucose (n=11), HDL and Non-HDL (n=10), triglycerides (n=8), and LDL (n=7). Fasting total cholesterol (p = 0.034, d = -0.22) and glucose (p = 0.001, d = -1.22) were greater before compared with after the

12 week exercise intervention. There were no differences between baseline and post-intervention values for HDL (p = 0.146, d = -0.40), non-HDL (p = 0.856, d = -0.02), LDL (p = 0.652, d = 0.11) and triglycerides (p = 0.776, d = -0.09).

Table 6.4 Participants' metabolic profile

	Baseline	After intervention
Total cholesterol (mmol/L)*	4.30 ± 0.94	4.10 ± 1.02
HDL (mmol/L)	1.18 ± 0.29	1.08 ± 0.25
Non-HDL (mmol/L)	3.19 ± 0.89	3.17 ± 0.93
LDL (mmol/L)	2.98 ± 0.71	3.05 ± 0.69
Triglycerides (mmol/L)	1.01 ± 0.46	0.97 ± 0.38
Glucose (mmol/L)**	5.01 ± 0.35	4.58 ± 0.38

N=11 for Total cholesterol and glucose, N=10 for HDL and Non-HDL, N=8 for triglycerides and N=7 for LDL; HDL = high density lipoproteins; values presented as mean ± SD; Non-HDL = non-high density lipoproteins; LDL = low density lipoproteins. * Means significantly different ($p < 0.05$); ** Means significantly different ($p < 0.01$).

6.3.6 Eating behaviour and food cravings

Scores for cognitive restraint, uncontrolled eating, emotional eating, total and specific food cravings are summarised in table 6.5. There were no differences between baseline and post-exercise intervention scores for cognitive restraint, uncontrolled eating and emotional eating scores ($p > 0.05$). However, there was a trend (p = 0.058, d = 0.68) for cognitive restraint to be greater after the exercise intervention than at baseline. Total food cravings (p = 0.009, d = -1.19) and specific cravings of high-fat foods (p = 0.023, d

= -0.90), fast-food fats (p = 0.009, d = -0.71) and carbohydrates/starches (p = 0.009, d = -0.56) decreased from baseline to 12 weeks. There was also a trend for cravings of sweets (p = 0.052, d = -0.86) to be lower after the exercise intervention compared to baseline.

Table 6.5 Eating behaviour and food cravings scores

	Baseline	**After intervention**
TFEQ-R18 scores (%)		
Cognitive restraint	24.7 ± 11.5	33.8 ± 16.0
Uncontrolled eating	41.3 ± 17.0	36.0 ± 15.4
Emotional eating	25.3 ± 23.4	26.3 ± 29.5
FCI scores (1-5 Likert scale)		
Total food cravings**	2.3 ± 0.4	1.9 ± 0.4
High-fats*	1.9 ± 0.5	1.5 ± 0.4
Fast-food fats**	2.9 ± 0.7	2.4 ± 0.8
Carbohydrate/starches**	2.3 ± 0.8	1.9 ± 0.7
Sweets	2.3 ± 0.6	1.9 ± 0.4

N=11; values presented as mean ± SD; TFEQ-R18 = revised version of the three-factor eating questionnaire; FCI= food cravings inventory; * Means significantly different (p < 0.05); ** Means significantly different (p < 0.01).

6.3.7 Energy and macronutrient intake

Daily energy intakes are shown in figure 6.6. There were no intervention (p = 0.326), time (p = 0.738) or interaction (p = 0.780) effects for energy intake. Similarly, there were no main or interaction effects for protein, fat and carbohydrate intake (p > 0.05) (Table 6.6).

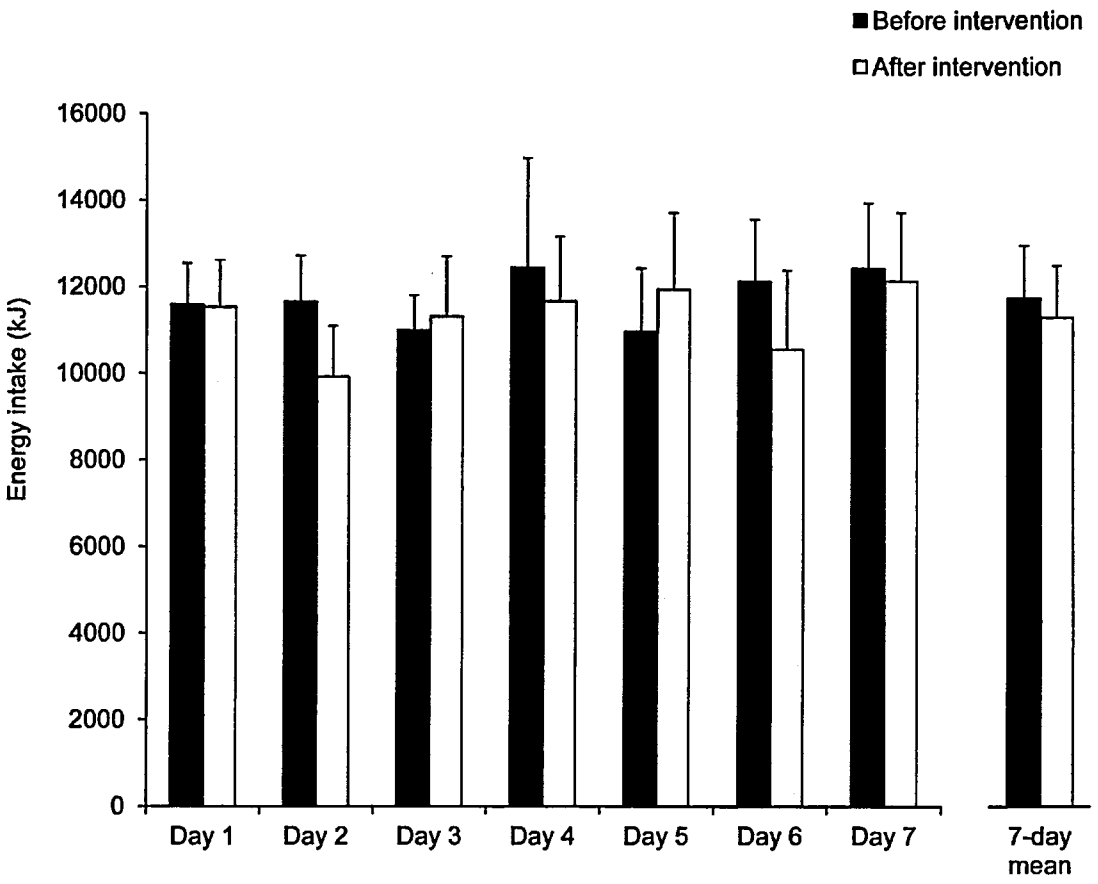


Figure 6.6 Daily and 7-day mean energy intake before and after the 12 week exercise intervention (n=11; means ± SEM).

Table 6.6 Mean 7-day macronutrient intake

	Baseline	After intervention
Protein intake (%)	15.9 ± 2.3	17.1 ± 3.0
Fat intake (%)	32.3 ± 4.8	33.1 ± 6.2
Carbohydrate intake (%)	51.8 ± 5.2	49.8 ± 6.7

N=11; values presented as mean ± SD.

6.3.8 Energy expenditure

Three participants did not have complete 7-day Actiheart data for at least one of the two measurement periods, therefore analyses were made for 9 participants. There were no intervention (p = 0.799), time (p = 0.765) or interaction (p = 0.696) effects for energy expenditure. Likewise there was no main or interaction effects for physical activity energy expenditure ($p > 0.05$) (Figure 6.7).

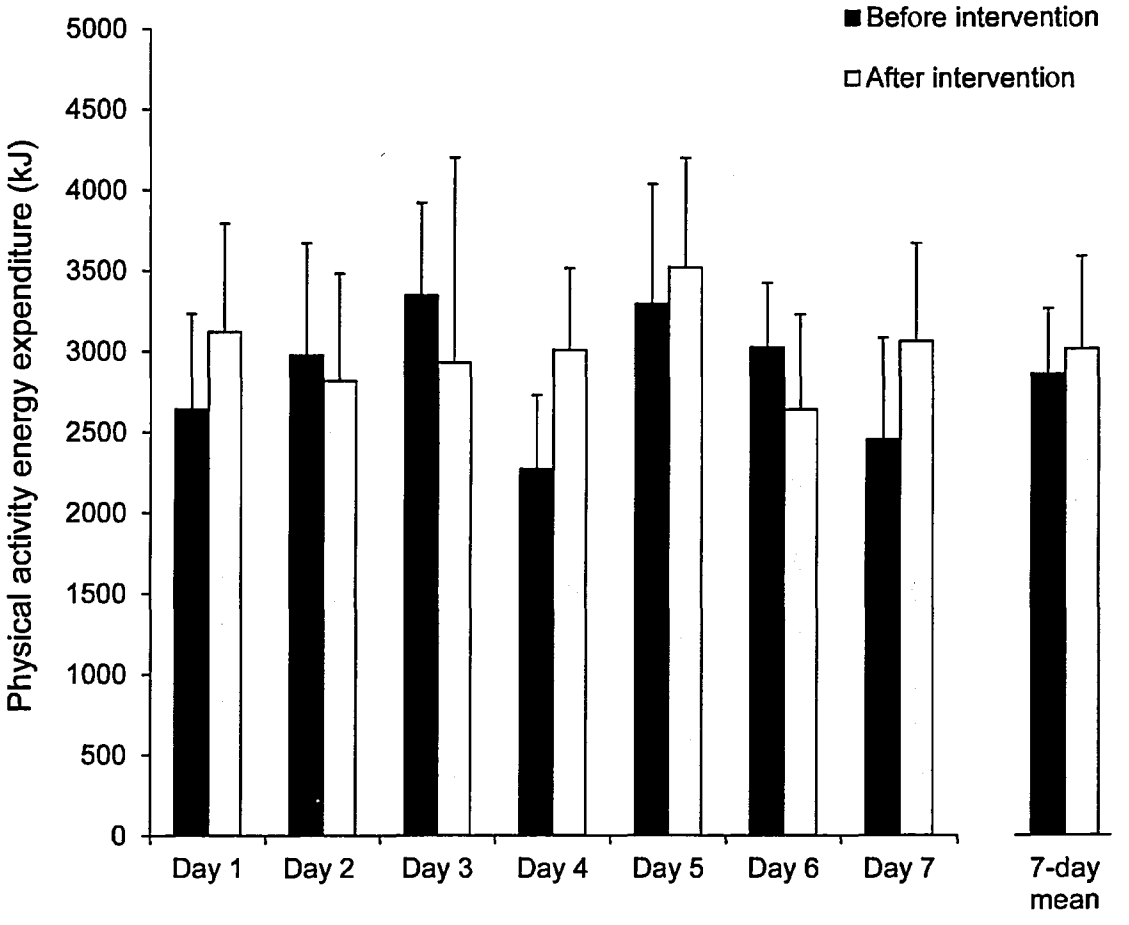


Figure 6.7 Daily and 7-day mean physical activity energy expenditure before and after the 12 week exercise intervention (N=9; means ± SEM).

6.4 Discussion

The main finding arising from this study is that 12 weeks of moderate-intensity aerobic exercise does not elicit changes in 7-day energy intake or expenditure in inactive men. In addition, the lack of difference between pre- and post-exercise intervention energy expenditure suggest that participants in this study reduced their activity levels to values similar to baseline immediately after direct supervision and support was no longer available.

In the present study, participants' mean maximum oxygen consumption at baseline was within the normal range for untrained men (Saltin & Astrand, 1967), however, the values were still greater than anticipated based on the directly measured maximum oxygen consumption values of the group of inactive men presented in chapter 4. This difference can be explained by the Astrand-Ryhming submaximal cycle ergometer test having an error of ±15% for untrained individuals (Macsween, 2001), which, as previously discussed in section 3.8.3, is due to the test underlying assumptions that there is a linear relationship between heart rate and oxygen consumption, maximum heart rate at a given age is constant and mechanical efficiency (oxygen consumption at a given exercise intensity) is the same for everyone.

Consistent with previous research (Broeder *et al.*, 1992; Caudwell *et al.*, 2013a; Caudwell *et al.*, 2013b), the twelve weeks exercise intervention delivered in this study induced changes in body composition. The reductions of mean body mass (-1.6 kg) and mean body fat (-1.1 kg) were modest and did not reach the minimum change (5% of body mass, i.e. mean change of 4 kg in this sample) necessary to reduce metabolic and cardiovascular disease risk (Blackburn, 1995; Wing *et al.*, 2011). However, this finding

can be explained by the moderate-intensity (60% HRR) and frequency (3 to 4 times per week) of the exercise sessions, the length of the exercise intervention (12 weeks), and only five participants being classified as overweight or obese at baseline. Participants mean value for waist circumference at baseline was not indicative of increased health risk (> 94 cm) (Han *et al.*, 1995) even when considering the specific optimal waist circumference cut-point of 87 cm for a body mass index between 18.5 and 24.9 in men (Ardern *et al.*, 2004). Nevertheless, the reduction in waist circumference (-3.8 cm) is important because of the association between excess abdominal obesity and increased risk of mortality, cardiovascular disease, diabetes, insulin resistance and metabolic syndrome (Katzmarzyk *et al.*, 2006; Klein *et al.*, 2007). Moreover, 12 weeks of moderate-intensity exercise made other positive changes in health markers such as the increase in the estimated maximum oxygen consumption and decrease in resting diastolic blood pressure, total cholesterol and fasting glucose. Together, these findings reinforce the health benefits of exercising even if reductions in body mass are modest.

The 12 week exercise intervention in this study produced a large inter-individual variability in changes of body and fat mass, a finding observed in several studies examining the effects of a 12 week supervised exercise intervention in overweight and obese men and women (Caudwell *et al.*, 2009; Caudwell *et al.*, 2013b; King *et al.*, 2008; King et al., 2009). In the present study, nine participants experienced various degrees of reduction in body and fat mass, one participant did not have any changes in body composition and one participant had a substantial increase in body and fat mass suggesting that compensatory responses may also be highly individual. Nevertheless, findings for the two latter participants are possibly explained by differences in the exercise intervention as the participant which did not have any changes in body

composition consistently reduced the intensity of the prescribed exercise for the unsupervised sessions (estimated exercise mean energy expenditure of supervised sessions = 2316 kJ and unsupervised sessions = 1527 kJ) while the participant with increases in body and fat mass had the highest percentage of unsupervised sessions (92%). This meant that most of the exercise sessions information was self-reported and therefore it is possible that these reports were not accurate. The variability of responses in the remaining nine participants could not be explained by differences in the amount of supervised and unsupervised sessions or estimated exercise energy expenditure. Therefore, it is possible that compensatory responses occurred but were not detected in the present study because of its small sample size and limited statistical power.

No changes were observed in the three-factor eating scores for cognitive restraint, uncontrolled eating and emotional eating after the 12 week exercise intervention, however, there was a tendency for participants to have a greater cognitive restraint score after the exercise intervention compared to baseline. This finding is perhaps unsurprising as increases in cognitive restraint have been associated with treatments inducing reductions in body mass (Foster *et al.*, 1998) such as in exercise (King *et al.*, 2009) and dietary (Westerterp-Platenga *et al.*, 1998) interventions. Previous findings make it unclear if this increase in cognitive restraint is due to the treatments or the reductions in body mass. However, it is possible that by being involved in interventions that aim to improve their health, participants' become more aware of their lifestyle, which in turn could increase their control over it. Anecdotally, many participants described their main motivation to be involved in the study as wanting to become healthier and/or losing body mass so it is possible that this motivation together with the exercise itself exerted an increase in their cognitive control over food. In contrast, the

total food cravings and specific cravings of high-fats, fast-food fats and carbohydrates/starches scores decreased from baseline to after the exercise intervention. These findings are in agreement with a recent study suggesting that chronic exercise training is associated with an attenuated response to visual food cues in brain regions known to be important in food intake regulation (Cornier *et al.*, 2012). Additionally, food cravings have previously been reported to be closely associated with mood, in particular as an antecedent and as a consequence of the food cravings (Hill *et al.*, 1991). Therefore, it is possible that the decrease in food cravings is related to the exercise-induced improvement in mood observed in this study (data not presented). To the authors knowledge no study has previously measured general and specific food cravings before and after an exercise intervention, however, reductions in food cravings have been previously reported in studies examining the effects of 12 weeks of a low-calorie and very-low calorie diets on food cravings (Harvey *et al.*, 1993; Martin *et al.*, 2006). These findings are not universal as Wadden *et al.* (1997) reported no differences on intensity of food cravings after 48 weeks of diet alone, diet plus aerobic training, diet plus strength training, or diet combined with aerobic and strength training. Additionally, Foster *et al.* (1992) did not find any effects of 24 weeks of three very-low-calorie diets on food cravings, however differences between studies results may be explained by these two later studies not using a validated measure of food cravings.

Similar to previous studies (Broeder *et al.*, 1992; Caudwell *et al.*, 2013a; Caudwell *et al.*, 2013b; Church *et al.*, 2009; Hollowell *et al.*, 2009; Keytel *et al.*, 2001; Turner *et al.*, 2010; Van Etten *et al.*, 1997) there were no changes in energy intake and energy expenditure after the exercise intervention. However, as previously discussed it is possible that compensatory responses were present but were not detected due to the

small sample size and limited statistical power. Of interest is that the energy expenditure after 12 weeks was similar to baseline, which suggests that most participants did not continue exercising for the same frequency and intensity after the end of the study exercise intervention. This is perhaps surprising because it occurred despite participants being offered 3 months free gym membership and encouraged to continue their exercise programme independently. This finding would explain why exercise training studies have not shown a consistent effect on long-term maintenance of body mass (Fogelholm & Kukkonen-Harjula, 2000; Franz *et al.*, 2007).

In summary, this study demonstrated that 12 weeks of moderate-intensity aerobic exercise does not induce changes on weekly energy intake and expenditure in inactive men at a group level. The inter-individual variability in changes in body and fat mass that could not be explained by differences in the exercise programme suggests a presence of compensatory responses that were possibly not detected due to the low sample size and limited statistical power of the present study. However, individual level analysis should be interpreted with caution as despite allowing a deeper and richer look at each individual there is limited control over error rates and conclusions are not generalizable. Findings from this study also reinforce the importance of exercising for health improvements even when reductions in body mass are modest. Moreover, data from this study suggest that some inactive men may not maintain the same volume of exercise without direct supervision even when having free access to a gym. The use of other strategies such as behavioural therapy may be needed to help inactive men maintain their exercise levels after the end of the exercise intervention.

Chapter 7: General discussion

7.1 Introduction

Appetite and physical activity are investigated because of their influence on energy balance. Previous research has highlighted the complexity of the psychobiological systems involved in their control (Blundell, 2011) and examined the influence of factors such as exercise (Jokisch *et al.*, 2012; King *et al.*, 2008; Unick *et al.*, 2010). Current knowledge supports that an acute bout of moderate-intensity exercise does not affect hunger or subsequent energy intake (Martins *et al.*, 2008). The research on the effects of an acute bout of exercise on non-exercise physical activity is limited but suggests that higher exercise intensities elicit compensatory changes in physical activity (Alahmadi *et al.*, 2011). Chronic exercise has been reported to induce a partial compensatory response in energy intake in the short-term (7 to 14 days) (Staten, 1991; Stubbs *et al.*, 2004a; Stubbs *et al.*, 2004b; Whybrow *et al.*, 2008), however, these responses have not been observed over longer periods of time (Caudwell *et al.*, 2013a; Caudwell *et al.*, 2013b; Donnelly *et al.*, 2003; Martins *et al.*, 2007b). In contrast, the effects of chronic exercise on physical activity energy expenditure are equivocal (Colley *et al.*, 2010; Hunter *et al.*, 2000; Rosenkilde *et al.*, 2012; Turner *et al.*, 2010).

The research presented within this thesis complements current knowledge on the acute and chronic effects of exercise on appetite and energy expenditure, and provides novel evidence about how these effects differ according to participant's physical activity and sex. This chapter will integrate the findings from all the studies presented within this thesis. The summary of each study design, measurements and findings is presented in table 7.1

Table 7.1 Summary of thesis studies

7.2 Hunger

Several studies have reported a transient suppression of appetite during high-intensity (Broom *et al.,* 2009; Broom et al., 2007; King *et al.,* 2013) but not during low- and moderate-intensity exercise (Jokisch *et al.,* 2012; King et al., 1994; Thompson *et al.,* 1988) in men. Evidence suggests that hunger returns to control values shortly after exercise (King & Blundell, 1995; King *et al.,* 1994) and remains the same during subsequent hours, even in the presence of large exercise-induced energy deficits (King *et al.,* 2010a). This suppressive effect of high-intensity exercise on hunger, termed "exercise-induced anorexia", is therefore believed to be dependent on the intensity of the exercise bout (Martins *et al.,* 2008). However, this observed relationship between exercise intensity and hunger has not been consistently reported in women (King *et al.,* 1996; Maraki *et al.,* 2005; Pomerleau *et al.,* 2004) suggesting that exercise may not suppress hunger in the same way for men and women. Therefore, the cross-sectional studies presented in this thesis examined the effects of 60 minutes of moderate-intensity exercise (50% of V02max) on subjective ratings of hunger in active and inactive men and women. In particular, the use of the same protocol in all cross-sectional studies is important as it allows a direct comparison of results from the three studies.

In study one, 60 min of moderate-intensity cycling in active (51.0 ± 3.8% of V02max; 1979 ± 266 kJ) and inactive (51.4 ± 3.4% of V02max; 1748 ± 206 kJ) men had no effect on hunger. Sixty minutes of moderate-intensity cycling in active (50.1 ± 2.1% of V 02max; 1462 ± 157 kJ) and inactive (55.2 ± 9.5% of V02max; 1127 ± 275 kJ) women not using hormonal contraceptives did not elicit an effect on hunger in study two. Likewise, 60 min of moderate-intensity cycling in active (51.2 ± 2.2% of VCEmax; 1387 ± 139 kJ)

and inactive (54.0 ± 7.5% of $\dot{V}O_{2max}$; 1304 ± 248 kJ) women taking oral contraceptives did not have an effect on hunger in study three. Therefore, findings from the cross-sectional studies of this thesis suggest that 60 minutes of moderate-intensity cycling has no effect on hunger irrespectively of sex or habitual physical activity. These findings are in agreement with previous evidence in men supporting that moderate-intensity exercise is not sufficient to induce "exercise-induced anorexia" (Jokisch *et al.*, 2012; King *et al.*, 1997; King *et al.*, 2010b) and provides evidence that, under the same conditions, pre-menopausal women respond similarly.

The difference in the intensity of hunger ratings irrespective of condition or participants' habitual physical activity between study two and three was also examined. A three-way repeated measures ANOVA (study x condition x time) demonstrated that women taking oral contraceptives had consistently higher hunger ratings during both experimental days compared with women not using hormonal contraceptives (mean difference = 8 mm; p = 0.049). Considering that there were no differences between participants baseline characteristics (age, stature, body mass, maximum oxygen consumption) (p > 0.05), mean dietary restraint scores (p > 0.05) and mean premenstrual symptoms (p > 0.05) between the two studies, it is possible that these findings are due to the use of oral contraceptives. To the authors knowledge only one study (Hagobian *et al.*, 2012) examining the effects of exercise on appetite and energy intake reported that women were taking OCs, however comparisons with this study are not possible because hunger data were not presented. Nevertheless, oral contraceptives have previously been demonstrated to decrease the basal level of serum cholecystokinin (CCK), a hormone known for its appetite-suppressing effect, in young healthy women (Hirschberg *et al.*, 1996; Karlsson *et al.*, 1992; Naessen *et al.*, 2007). Oestrogen produced in the stomach

has also been demonstrated to directly induce ghrelin (appetite-stimulating hormone) expression and production in both female and male rat stomachs (Matsubara *et al.,* 2004; Sakata *et al.,* 2006). When examined in young women no differences were observed between fasting levels of total ghrelin in users and non-users of oral contraceptives (St-Pierre *et al.,* 2006). However, ghrelin circulates in two forms (acylated and des-acylated) (Hosoda *et al.,* 2000), and acylation of ghrelin is believed to be essential for its appetite-stimulatory effects due to being able to cross the blood-brain barrier (Murphy & Bloom, 2006). Thus it is possible that changes in acylated ghrelin were not detected. Collectively, there is some evidence suggesting that the use of oral contraceptives has an effect on appetite, therefore it would be of interest for future research to directly examine the effects of oral contraceptives on appetite-regulating hormones such as acylated ghrelin and peptide YY$_{3.36}$.

7.3 Ad libitum lunch energy intake

Most of the previous research suggests that an acute bout of exercise has no effect on the first meal after exercise in active men (Deighton *et al.,* 2012; Kelly *et al.,* 2012; Vantansever-Ozen *et al.,* 2011) and women (Finlayson *et al.,* 2009; Hagobian *et al.,* 2012; Larson-Meyer *et al.,* 2012). This lack of an effect is also observed in most studies examining inactive-to-moderately-active women (George & Morganstein, 2003; Maraki *et al.,* 2005; Unick *et al.,* 2010), but not men (Jokisch, *et al.,* 2012; Ueda *et al.,* 2009b). As previously discussed, different research designs and methodological limitations make interpretation of findings difficult, therefore the cross-sectional studies presented in this thesis aimed to, under the same conditions, examine the effects of an acute bout

of moderate-intensity exercise on energy intake while controlling for participants' physical activity and sex.

In study one, 60 min of moderate-intensity cycling did not change energy intake at lunch in active men and exerted a non-significant increase (432 kJ) in inactive men. The percentage of energy compensation at lunch was lower in the active group (14%) than the inactive (30%). When accounting for the energy expended during the 60 minutes of exercise/rest, relative energy intake in the exercise condition was lower than the control condition for both groups. This meant that after lunch active participants were in a greater energy deficit (-1596 kJ) than inactive participants (-885 kJ), a finding that can be explained by the combined increase in energy intake by inactive participants (432 kJ) and the higher energy expended during the 60 min of exercise by active participants (231 kJ).

Similarly, in study two, moderate-intensity cycling did not elicit any changes and induced a non-significant increase (425 kJ) in energy intake at lunch in active and inactive women not using hormonal contraceptives, respectively. In this study, active and inactive participants compensated for 13% and 47% of the net exercise-induced energy expenditure, respectively. Relative energy intake decreased after exercise compared to control only in active individuals thus as in the previous study with men active participants were in a greater energy deficit (-1023 kJ) than inactive participants (-392 kJ) after lunch. This finding is explained by the higher energy expended during the 60 min of exercise by active participants (335 kJ) and the changes in the energy intake at lunch.

When examining the effects of an acute bout of moderate-intensity cycling in women taking oral contraceptives (study three) there was an overall condition effect with a greater lunch energy intake after exercise than control (328 kJ). However, a separate analysis demonstrated a trend for a greater energy intake after exercise (437 kJ) only in the active group. In contrast with the previous two studies where inactive participants had a higher percentage of energy compensation at lunch, in this study, active participants had a higher percentage of energy compensation (43%) compared to the inactive group (16%). Despite this tendency for a higher energy intake after exercise, relative energy intake was reduced in the exercise compared with the control condition in both groups. In contrast with the previous two studies the energy deficit of this study active (-641 kJ) and inactive (-742 kJ) participants after lunch was similar.

Findings from the cross-sectional studies of this thesis suggest that the effect of 60 min of moderate-intensity cycling on the *ad libitum* energy intake may be dependent on participants' habitual physical activity and, in women, on the use of oral contraceptives. The first two studies demonstrated a tendency for inactive participants to increase their energy intake whereas active participants remained approximately the same. However, this was not the case when examining women taking oral contraceptives (study three), where active but not inactive participants increased their post-exercise lunch energy intake. While findings from studies one and two can be explained by a psychological drive in inactive participants to use food as a reward for exercising (King *et al.,* 2007), the third cannot. Considering that the main identifiable difference between the two studies with women was the use of oral contraceptives it is possible that these interfere with the short-term (i.e. episodic) signals associated with meal termination (i.e. satiation) (Blundell *et al.,* 2008). An increase in perceived palatability of food could

also explain these findings as this has previously been reported in active lean women who are prone to increase their energy intake (Finlayson *et al.*, 2009), however this study did not report if participants took oral contraceptives. It is important to note that these studies were not designed to detect the underlying mechanisms of these effects and therefore future research should examine the effects of an acute bout of exercise in these populations with measurements of physiological markers such as appetite-regulating hormones, and the use of qualitative approaches to confirm if psychological drives are present.

As mentioned in the previous section, there were no differences between participants' baseline characteristics in studies two and three and therefore it is possible that differences between these studies are due to the use of oral contraceptives. Indeed, as observed with hunger, two-way repeated measures ANOVAs (study x condition) demonstrated that participants who were taking oral contraceptives had an overall greater energy intake (mean difference = 625 kJ; $p = 0.022$) and relative energy intake at lunch (mean difference = 596 kJ; $p = 0.025$). It is possible that this greater energy intake is associated to the decreased secretion of CCK (in response to a meal) observed in women 3 months after starting to take oral contraceptives (Naessen *et al.*, 2007).

The low sample size and associated lack of statistical power in studies two and three mean that these findings should be interpreted with caution. Nevertheless, the well-controlled design, concurrent interpretation of the results of energy intake at lunch, the percentages of energy compensation, the energy deficits after lunch, and the confirmation of a condition effect for energy intake ($p = 0.014$) when data from the two studies is analysed together gives more confidence to previous interpretations.

7.4 Free-living energy intake

From the few studies that have examined the effects of an acute bout of exercise on subsequent free-living energy intake most have been limited to an observation period of 24 hours (Jokisch *et al.*, 2012; Klausen *et al.*, 1999; Lluch *et al.*, 1998; Maraki *et al.*, 2005). Longer observation periods could reveal an exercise-induced compensation in energy intake at subsequent eating episodes. Therefore, the cross-sectional studies of this thesis examined the effects of 60 min of moderate-intensity cycling on free-living energy intake during the remainder of the experimental day and subsequent three days. Additionally, the high variability observed in exercise-induced reductions in body mass and fat tissue are believed to be partly due to compensatory responses (King *et al.*, 2007). Thus the last study of this thesis (study four) focused on examining the effects of 12 weeks of moderate-intensity aerobic exercise on 7-day free-living energy intake in inactive men. As previously mentioned, this target population was selected due to physical inactivity being a major risk factor for the development of diseases such as obesity (Haskell *et al.*, 2009), and the difficulties of controlling the variables such as menstrual cycle, premenstrual symptoms and use of hormonal contraceptives.

In study one, 60 min of moderate-intensity cycling induced an increase in active participant's free-living energy intake during the remainder of the exercise experimental day compared with control, but no changes were observed in energy intake over the subsequent three days. In contrast, inactive participants only increased their energy intake on the third day after exercise experimental day compared with control. Percentages of energy compensation demonstrated that active participants compensated for 127% by the end of the experimental day whereas inactive participants compensated for 172% only on the third day after the experimental day. In contrast, study two did not

demonstrate any changes in free-living energy intake after the exercise experimental day compared with control. However, percentages of energy compensation suggest a possible delayed compensation on the second day after the experimental day (137%) in inactive participants. In study three, there were no differences in free-living energy intake during the remainder of the exercise experimental day compared with control in active participants but inactive decreased their energy intake on the first day after the experimental day having a compensation of -176% on that day.

The cross-sectional studies of this thesis suggest that, in men, 60 min of moderate-intensity cycling is able to induce acute and delayed compensatory responses in energy intake dependent on participants' habitual physical activity. Moreover, the incomplete compensation observed in both groups at the end of day four gives further support to the use of exercise as a strategy to induce short-term energy deficits. In women this relationship could not be confirmed despite the percentages of energy compensation giving some indication of possible differences. These findings are possibly explained by the low sample size in studies two and three, which meant that these studies had limited statistical power to detect changes in the free-living period of the studies and therefore more experimental studies are necessary to clarify and confirm these outcomes. Additionally, a three-way repeated measures ANOVA (study x condition x time) demonstrated that participants who were taking oral contraceptives and not using hormonal contraceptives had a similar energy intake over the 4 days (mean difference = 123 kJ; p = 0.375). This finding is somewhat surprising considering differences observed during the laboratory period. The lack of statistical power of these studies, the different expectations and behaviour under controlled conditions, or some degree of underreporting in the free-living energy intake data are possible explanations for these

findings. To examine the latter possibility, analyses were conducted after the exclusion of participants with a mean ratio of energy intake to basal metabolic rate lower than Goldberg *et al.* (1991) cut-off of 1.1. Nevertheless, findings from the study effect of this three-way repeated measures ANOVA (study x condition x time) remained similar (mean difference = 132 kJ; $p = 0.320$).

Having examined the effects of an acute bout of moderate-intensity exercise in studies one, two and three, the focus of this thesis moved towards the investigation of the chronic effects of 12 weeks of moderate-intensity exercise on 7-day free-living energy intake. In study four, no changes were observed on weekly energy intake from baseline to after the exercise intervention, however, the high inter-individual variability in body and fat mass changes suggested that some compensatory responses may have occurred. These findings can be explained by the low sample size and limited statistical power of this study therefore further research is needed to identify these responses. Nevertheless, results of several health markers measured in this study reinforces the importance of exercising for health improvements even if only eliciting modest decreases in body mass.

7.5 Free-living energy expenditure

To the author's knowledge only one study has examined the effects of a single bout of exercise on non-exercise physical activity in adults (Alahmadi *et al.*, 2011), and there are equivocal findings from several studies examining the effect of chronic exercise (Colley et al., 2010; Rosenkilde *et al.*, 2012; Turner *et al.*, 2010). Therefore the cross-sectional studies (studies one, two and three) presented in this thesis aimed to, under the same conditions, examine the effects of an acute bout of moderate-intensity cycling on

free-living energy expenditure during the remainder of the experimental day and subsequent three days while controlling participants' physical activity and sex. Additionally, study four aimed to investigate the effects of 12 weeks of moderate-intensity aerobic exercise on 7-day free-living energy expenditure in inactive men.

No differences were observed in free-living physical activity energy expenditure after exercise compared with control in study one suggesting that irrespective of participants' habitual physical activity an acute bout of moderate-intensity cycling does not elicit compensatory changes in physical activity in men. These findings agree with previous research in adults that reported an increase in non-exercise physical activity, after a high- but not moderate-intensity exercise session (Alahmadi *et al.*, 2011), suggesting that these effects may be dependent on the intensity of exercise. Similarly no changes were observed in studies two and three, however these studies were not sufficiently powered to detect changes in the free-living period of the study. For instance, visual analysis of studies two and three daily physical activity energy expenditure data (Figure 5.6; Figure 5.11) suggest that in study two, active participants may have increased their daily physical activity during the three days after the exercise experimental chapter compared to control, whereas in study three the opposite was observed. Moreover, in both studies the inactive group seemed to increase their energy intake on the second day after the exercise experimental day. An analysis of both studies data with a four-way repeated measures ANOVA (study x group x condition x time) demonstrated a condition x study interaction (p = 0.049) suggesting that participants that did not use hormonal contraceptives increased their energy expenditure over the three days after the exercise experimental day (mean difference = 701 kJ) whereas participants taking oral contraceptives reduced it (mean difference = -237 kJ).

Collectively these findings give support to a possible exercise-induced compensatory response in energy expenditure dependent on the use of hormonal contraceptives, however, future research with adequate sample sizes are needed to confirm this.

In study four, 12 weeks of moderate-intensity aerobic exercise did not induce changes on weekly expenditure in inactive men. Indeed, the mean 7-day physical activity energy expenditure after the exercise intervention was very similar to baseline (mean difference = + 156 kJ). This was unexpected because it occurred despite participants being offered a 3 months free gym membership and encouraged to continue their exercise programme independently. It is possible that these participants relied on extrinsic motivation to exercise and as soon as this factor was not present they reduced substantially their activity due to their lower intrinsic motivation to exercise. This is supported by anecdotal evidence as many participants described their main motivation to be involved in the study as wanting to become healthier and/or losing body mass and lacking the motivation to do it on their own, stating that they required someone to "make them exercise". It is also possible that the extrinsic motivation provided by the researcher decreased their intrinsic motivation as according to the Cognitive Evaluation Theory if recipients of extrinsic "rewards" (e.g. praise) interpret these as indicating external control this can decrease their feelings of self-control and therefore undermine their intrinsic motivation (Deci & Ryan, 1985). Moreover, the loss of this extrinsic motivation may have also affected their enjoyment of exercise as the associated social interaction was not present. Therefore, exercise interventions aiming to increase participant's physical activity and sustain those levels after the intervention should aim to include other strategies that can possibly make this transition easier (e.g. behavioural therapy).

7.6 Limitations and recommendations for future research

The studies presented in this thesis have limitations and some of these could be addressed in future work in this area. Study participants were mostly young, lean and healthy men (studies one and four) and women (studies two and three), therefore findings might not apply to older or obese adults. Future studies should examine the effect of acute and chronic exercise on appetite, energy intake and expenditure in different population groups.

In the cross-sectional studies, exercise intensity was individually prescribed to reflect fitness status with both groups cycling at approximately the same relative intensity. This adjustment was important because persons differing in fitness respond in markedly different ways to an exercise challenge set at a fixed absolute intensity (Howley, 2001). Nevertheless, this meant that active participants expended more energy during exercise than inactive participants. As the exercise-induced energy expenditure is a likely factor to induce energy compensation future studies should examine the effects of similar exercise-induced energy expenditures.

The control of the time of the testing (i.e. phase of the menstrual cycle) in studies two and three means that findings from these studies are possibly limited to the mechanisms operating at the examined stages. Moreover, the third study examined women taking different oral contraceptives and therefore it is not possible to draw any conclusions on the effects of specific oral contraceptive formulations. Future studies should conduct testing at different stages of the menstrual cycle and if possible examine specific combinations of oral contraceptives.

Energy intake is affected by other factors that could not be controlled in the free-living so it may be that observed differences in energy intake in studies 1 and 3 did not arise from physiological regulatory mechanisms but from behavioural/psychological (e.g. emotional states) and/or environmental factors (e.g. presence of other people at meal times). Similar studies are needed to confirm these findings and explore potential factors mediating the findings. The latter could possibly be achieved by including the measurement of physiological factors such as appetite-regulating hormones and using qualitative methods to identify behaviour/psychological and environmental factors.

The collection of energy intake and expenditure data in the free-living is highly dependent on participant compliance with methods and instructions making it more susceptible to errors in data collection. Therefore there is a need to develop new methods and explore the validity and reliability of possible combinations of current methods. In particular, self-reported food diaries are prone to bias, usually towards under-reporting of energy and misreporting of macronutrients (Livingstone & Black, 2003). To identify possible underreporting, the mean ratio of energy intake to basal metabolic rate was calculated for participants in all studies:

Study one:

- Active group (exercise vs. control: 1.6 ± 0.25 vs. 1.58 ± 0.3).
- Inactive participants (exercise vs. control: 1.41 ± 0.37 vs. 1.42 ± 0.33).
- Individual analysis showed that two inactive participants had a ratio lower than Goldberg *et al.* (1991) cut-off of 1.1 in one of the conditions (participant 13 exercise condition = 1.08; participant 21 control condition = 1.06).

Study two:

- Active group (exercise vs. control: 1.51 ± 0.26 vs. 1.50 ± 0.44).
- Inactive participants (exercise vs. control: 1.23 ± 0.24 vs. 1.31 ± 0.42).
- Individual analysis showed that two participants in the active group and one participant in the inactive group had a ratio lower than Goldberg *et al.* (1991) cut-off of 1.1 (participant 10 exercise condition = 1.01; participant 10 control condition = 0.92; participant 26 control condition = 0.99; participant 14 control condition = 0.76; participant 14 exercise condition = 0.71).

Study three:

- Active group (exercise vs. control: 1.39 ± 0.27 vs. 1.41 ± 0.13).
- Inactive participants (exercise vs. control: 1.67 ± 0.22 vs. 1.58 ± 0.25).
- Individual analysis showed that two participants in the active group had a ratio lower than Goldberg *et al.* (1991) cut-off of 1.1 (participant 14 control condition = 0.95; participant 24 control condition = 1.00).

Study four:

- All participants (baseline vs. after exercise intervention: 1.48 ± 0.32 vs. 1.44 ± 0.31).
- Individual analysis showed that no participants had a ratio lower than Goldberg *et al.* (1991) cut-off of 1.1.

A combination of the stringent inclusion criteria and the research design chosen to control for factors related with participants menstrual cycle meant that the second and

third studies (chapter 5) did not reach the estimated sample size. The high attrition rate of 30.4% in study 2 and 31% in study three is of concern and is attributable to the one-day choice that participants had for the second experimental days. As previously discussed, participants completed the two experimental days during days 5-9 of their menstrual cycle (study two) and during the first week they restarted taking the oral contraceptives or if continuous when a new pack was started (study three). As both experimental days were on the same day of the week this meant that in both studies the booking of the second experimental day was limited to one day per calendar month. For instance, if the first visit was completed on a Tuesday, the second experimental day had to be the first Tuesday after the 5th day of the participants' next menstrual cycle (study two) or the Tuesday of the week when participants restarted taking the pill (study three). This meant that if the participant, researcher or facilities were not available on that day a rescheduling for the next month was necessary. This led a total of seven (study two) and four participants (study three) to withdraw from the studies due to inability to book a second experimental day. From the 16 participants that completed study two only one participant from the active group completed the experimental days two months apart with all other participants completing the two experimental days in consecutive months. In study three, only two participants from the inactive group completed the experimental days two months apart with all other participants completing the two experimental days in consecutive months. Future studies examining the effects of exercise on energy intake and expenditure in women should consider adjustments in the inclusion criteria (e.g. allowing other forms of hormonal contraceptives such as implants and depot) and/or research design (e.g. increasing the period over which experimental days can be booked to the first 15 days of the menstrual cycle). It is also possible that additional resources (e.g. more researchers and incentives) may improve

attrition and recruitment rates. For example, the practical work of this thesis was conducted by a single researcher, which involved managing three studies simultaneously and therefore limiting the available time to reschedule experimental days. If a similar research design is employed it is necessary to increase the estimated sample size by at least 30% to account for dropouts.

As previously discussed, three of the studies presented in this thesis (studies two, three and four) had a lower than predicted sample size, therefore it is possible that the lack of an effect of exercise on energy intake and expenditure during the free-living period of these studies was due to lack of statistical power. Thus future studies should be powered appropriately to detect possible exercise-induced differences in free-living energy intake and expenditure.

The low sample size in study four can be explained by a combination of the demanding nature of the study and the strict inclusion criteria. From the 12 participants that started the study, one withdrew from the study because he did not want to follow the prescribed exercise intensities. A lack of a control group is also a limitation because it is not possible to conclude if findings are solely attributable to the exercise participation or other non-exercise related behaviour changes that may have occurred. Therefore future research should examine the effects of an exercise intervention with a randomised control trial design.

7.7 Conclusion

In conclusion, the research presented in this thesis provides a valuable insight into the effects of an acute bout of exercise on hunger, energy intake and expenditure in active and inactive men and women.

Specifically, this research shows that an acute bout of moderate-intensity aerobic exercise did not affect hunger irrespective of sex or habitual physical activity, however the use of oral contraceptives may have heightened appetite in women. Moreover, the effects of an acute bout of exercise on *ad libitum* energy intake may be dependent on participants' habitual physical activity and, in women, on the use of oral contraceptives. In men, exercise was also able to induce acute and delayed compensatory responses in energy intake dependent on participants' habitual physical activity. In women, no clear relationship was apparent but the percentages of energy compensation gave some indication of possible differences. In addition, an acute bout of exercise did not elicit compensatory changes in physical activity in men and, in women, any exercise-induced compensatory response in energy expenditure may have been dependent on the use of oral contraceptives.

This thesis provides evidence of the difficulties involved in designing well-controlled investigations in this field of study and in recruiting and retaining targeted populations. This research has also highlighted that irrespective of free access to exercising facilities, most inactive participants are not able to independently maintain their physical activity behaviour after the end of a supervised exercise intervention. Furthermore, this thesis lays the foundations for further work and development of appetite research controlling

important variables such as participants' habitual physical activity and the use of hormonal contraceptives.

References

Aarsland, A., Chinkes, D., & Wolfe, R. R. (1997). Hepatic and whole-body fat synthesis in humans during carbohydrate overfeeding. *The American Journal of Clinical Nutrition, 65(6),* 1774-1782.

Abbott, W., Howard, B. V., Christin, L., Freymond, D., Lillioja, S., Boyce, V. L., et al. (1988). Short-term energy balance: Relationship with protein, carbohydrate, and fat balances. *American Journal of Physiology-Endocrinology and Metabolism, 255(3),* E332-E337.

Abizaid, A., Liu, Z., Andrews, Z. B., Shanabrough, M., Borok, E., Elsworth, J. D., et al. (2006). Ghrelin modulates the activity and synaptic input organization of midbrain dopamine neurons while promoting appetite. *Journal of Clinical Investigation,* 776(12), 3229-3239.

Acheson, K., Schutz, Y., Bessard, T., Anantharaman, K., Flatt, J., & Jequier, E. (1988). Glycogen storage capacity and de novo lipogenesis during massive carbohydrate overfeeding in man. *The American Journal of Clinical Nutrition, 48(2),* 240-247.

Adriaanse, M. A., Vinkers, C. D., De Ridder, D. T., Hox, J. J., & De Wit, J. B. (2011). Do implementation intentions help to eat a healthy diet? A systematic review and meta-analysis of the empirical evidence. *Appetite,* 56(1), 183-193.

Ahima, R. S., & Antwi, D. A. (2008). Brain regulation of appetite and satiety. *Endocrinology and Metabolism Clinics of North America, 37(4),* 811-823.

Alahmadi, M., Hills, A. P., King, N. A., & Byrne, N. M. (2011). Exercise intensity influences NEAT in overweight and obese adults. *Medicine & Science in Sports & Exercise, 43(4),* 624-631.

Allen, S. S., McBride, C. M., & Pirie, P. L. (1991). The shortened premenstrual assessment form. *The Journal of Reproductive Medicine, 36*(11), 769-772.

Alpert, S. S. (1990). Growth, thermogenesis, and hyperphagia. *The American Journal of Clinical Nutrition, 52*(5), 784-792.

Anderson, E. S., Wojcik, J. R., Winett, R. A., & Williams, D. M. (2006). Social-cognitive determinants of physical activity: The influence of social support, self-efficacy, outcome expectations, and self-regulation among participants in a church-based health promotion study. *Health Psychology, 25*(4), 510-520.

Anderson, J. W., Konz, E. C., Frederich, R. C., & Wood, C. L. (2001). Long-term weight-loss maintenance: A meta-analysis of US studies. *The American Journal of Clinical Nutrition, 74*(5), 579-584.

Andre, D., & Wolf, D. L. (2007). Recent advances in free-living physical activity monitoring: A review. *Journal of Diabetes Science and Technology (Online), 1*(5), 760.

Anglé, S., Engblom, J., Eriksson, T., Kautiainen, S., Saha, M., Lindfors, P., et al. (2009). Three factor eating questionnaire-R18 as a measure of cognitive restraint, uncontrolled eating and emotional eating in a sample of young Finnish females. *International Journal of Behavioral Nutrition and Physical Activity, 17*(6), 41.

Ardern, C. I., Janssen, I., Ross, R., & Katzmarzyk, P. T. (2004). Development of health - related waist circumference thresholds within BMI categories. *Obesity Research, 12*(7), 1094-1103.

Astrand, I. (1960). Aerobic work capacity in men and women with special reference to age. *Acta Physiologica Scandinavica, 49*(suppl 169), 1-92.

Astrand, P., & Rhyming, I. (1954). A nomogram for calculation of aerobic capacity (physical fitness) from pulse rate during submaximal work. *Journal of Applied Physiology, 7*, 218-221.

Atkinson, G., & Nevill, A. M. (1998). Statistical methods for assessing measurement error (reliability) in variables relevant to sports medicine. *Sports Medicine, 26*(4), 217-238.

Atwater, W., & Rosa, E. (1899). A new respiration calorimeter and experiments on the conservation of energy in the human body, II. *Physical Review (Series I), 9*(4), 214.

Babicz-Zielińska, E. (2006). Role of psychological factors in food choice–a review. *Polish Journal of Food and Nutrition Sciences, 15*(4), 379-384

Balaguera-Cortes, L., Wallman, K. E., Fairchild, T. J., & Guelfi, K. J. (2011). Energy intake and appetite-related hormones following acute aerobic and resistance exercise. *Applied Physiology, Nutrition, and Metabolism, 36*(6), 958-966.

Bancroft, J., & Rennie, D. (1993). The impact of oral contraceptives on the experience of perimenstrual mood, clumsiness, food craving and other symptoms. *Journal of Psychosomatic Research, 37*(2), 195-202.

Barreira, T. V., Kang, M., Caputo, J. L., Farley, R. S., & Renfrow, M. S. (2009). Validation of the actiheart monitor for the measurement of physical activity. *International Journal of Exercise Science, 2*(1), 60-71.

Bauman, A. E., Sallis, J. F., Dzewaltowski, D. A., & Owen, N. (2002). Toward a better understanding of the influences on physical activity: The role of determinants, correlates, causal variables, mediators, moderators, and confounders. *American Journal of Preventive Medicine, 23*(2), 5-14.

Beksinska, M. E., Smit, J. A., Kleinschmidt, I., Milford, C., & Farley, T. M. M. (2010). Prospective study of weight change in new adolescent users of DMPA, NET-EN, COCs, nonusers and discontinuers of hormonal contraception. *Contraception, 81*(1), 30-34.

Bellisle, F., & Dalix, A. (2001). Cognitive restraint can be offset by distraction, leading to increased meal intake in women. *The American Journal of Clinical Nutrition, 74*(2), 197-200.

Bellisle, F., Dalix, A. M., Airinei, G., Hercberg, S., & Péneau, S. (2009). Influence of dietary restraint and environmental factors on meal size in normal-weight women. A laboratory study. *Appetite,53*(3), 309-313

Bellisle, F., Dalix, A., & Slama, G. (2004). Non food-related environmental stimuli induce increased meal intake in healthy women: Comparison of television viewing versus listening to a recorded story in laboratory settings. *Appetite, 43*(2), 175-180.

Benelam, B. (2009). Satiation, satiety and their effects on eating behaviour. *Nutrition Bulletin, 34*(2), 126-173.

Berenson, A. B., & Rahman, M. (2009). Changes in weight, total fat, percent body fat, and central-to-peripheral fat ratio associated with injectable and oral contraceptive use. *American Journal of Obstetrics and Gynecology, 200*(3), E1-E8.

Bertenshaw, E. J., Lluch, A., & Yeomans, M. R. (2009). Dose-dependent effects of beverage protein content upon short-term intake. *Appetite, 52*(3), 580-587.

Bertheussen, G. F., Romundstad, P. R., Landmark, T., Kaasa, S., Dale, O., & Helbostad, J. L. (2011). Associations between physical activity and physical and mental health–a HUNT 3 study. *Medicine & Science in Sports & Exercise, 43*(7), 1220-1228.

Berthoud, H. (2008). Vagal and hormonal gut–brain communication: From satiation to satisfaction. *Neurogastroenterology & Motility, 20*(s1), 64-72.

Berthoud, H. (2011). Metabolic and hedonic drives in the neural control of appetite: Who is the boss? *Current Opinion in Neurobiology, 21*(6), 888-896.

Berthoud, H., & Morrison, C. (2008). The brain, appetite, and obesity. *Annual Review of Psychology, 59*, 55-92.

Beyer, P. L., & Flynn, M. A. (1978). Effects of high- and low-fiber diets on human feces. *Journal of the American Dietetic Association, 72*(3), 271-277.

Bilski, J., Teleglów, A., Zahradnik-Bilska, J., Dembiński, A., & Warzecha, Z. (2009). Effects of exercise on appetite and food intake regulation. *Medicina Sportiva, 13*(2), 82-94.

Black, A., Goldberg, G., Jebb, S., Livingstone, M., Cole, T., & Prentice, A. (1991). Critical evaluation of energy intake data using fundamental principles of energy physiology: 2. evaluating the results of published surveys. *European Journal of Clinical Nutrition, 45*(12), 583.

Blackburn, G. (1995). Effect of degree of weight loss on health benefits. *Obesity Research, 3*(S2), 211s-216s.

Blundell, J. E. (2006). Perspective on the central control of appetite. *Obesity, 14 Suppl. 4*, 160S-163S.

Blundell, J. E. (2009). Exercise makes you fat–what's going on? *Nutrition Bulletin, 34*(4), 380-382.

Blundell, J. E. (2011). Physical activity and appetite control: can we close the energy gap?. *Nutrition Bulletin, 36*(3), 356-366.

Blundell, J. E., & King, N. A. (2000). Exercise, appetite control, and energy balance. *Nutrition, 16*(7-8), 519-522.

Blundell, J. E., Caudwell, P., Gibbons, C., Hopkins, M., Naslund, E., King, N., et al. (2012). Role of resting metabolic rate and energy expenditure in hunger and appetite control: A new formulation. *Disease Models & Mechanisms, 5*(5), 608-613.

Blundell, J. E., Stubbs, R. J., Hughes, D. A., Whybrow, S., & King, N. A. (2003). Cross talk between physical activity and appetite control: Does physical activity stimulate appetite? *Proceedings of the Nutrition Society, 62*(3), 651-661.

Blundell, J. E., De Graaf, C., Hulshof, T., Jebb, S., Livingstone, B., Lluch, A., et al. (2010). Appetite control: Methodological aspects of the evaluation of foods. *Obesity Reviews, 11*(3), 251-270.

Blundell, J. E., Levin, F., King, N. A., Barkeling, B., Gustafson, T., Hellstrom, P., et al. (2008). Overconsumption and obesity: Peptides and susceptibility to weight gain. *Regulatory Peptides, 149*(1), 32-38.

Booth, S. L., Sallis, J. F., Ritenbaugh, C., Hill, J. O., Birch, L. L., Frank, L. D., et al. (2001). Environmental and societal factors affect food choice and physical activity: Rationale, influences, and leverage points. *Nutrition Reviews, 59*(3), S21-S36.

Borg, G. A. V. (1973). Perceived exertion: A note on "history" and methods. *Medicine and Science in Sports, 5*(2), 90-93.

Boston, B. A., Blaydon, K. M., Varnerin, J., & Cone, R. D. (1997). Independent and additive effects of central POMC and leptin pathways on murine obesity. *Science, 278*(5343), 1641-1644.

Bowen, R. S., Turner, M. J., & Lightfoot, J. T. (2011). Sex hormone effects on physical activity levels. *Sports Medicine, 41*(1), 73-86.

Brage, S., Brage, N., Ekelund, U., Luan, J., Franks, P. W., Froberg, K., et al. (2006). Effect of combined movement and heart rate monitor placement on physical activity estimates during treadmill locomotion and free-living. *European Journal of Applied Physiology, 96*(5), 517-524.

Brage, S., Brage, N., Franks, P. W., Ekelund, U., Wong, M., Andersen, L. B., et al. (2004). Branched equation modeling of simultaneous accelerometry and heart rate monitoring improves estimate of directly measured physical activity energy expenditure. *Journal of Applied Physiology, 96*(1), 343-351.

Brage, S., Brage, N., Franks, P., Ekelund, U., & Wareham, N. (2005). Reliability and validity of the combined heart rate and movement sensor actiheart. *European Journal of Clinical Nutrition, 59(A),* 561-570.

Brage, S., Ekelund, U., Brage, N., Hennings, M. A., Froberg, K., Franks, P. W., et al. (2007). Hierarchy of individual calibration levels for heart rate and accelerometry to measure physical activity. *Journal ofApplied Physiology, 103(2),* 682-692.

Broeder, C., Burrhus, K., Svanevik, L., & Wilmore, J. (1992). The effects of either high-intensity resistance or endurance training on resting metabolic rate. *American Journal of Clinical Nutrition,* 55(4), 802.

Broom, D. R., Batterham, R. L., King, J. A., & Stensel, D. J. (2009). Influence of resistance and aerobic exercise on hunger, circulating levels of acylated ghrelin, and peptide YY in healthy males. *American Journal ofPhysiology - Regulatory, Integrative and Comparative Physiology, 296,* R29-R35.

Broom, D. R., Stensel, D. J., Bishop, N. C., Bums, S. F., & Miyashita, M. (2007). Exercise-induced suppression of acylated ghrelin in humans. *Journal of Applied Physiology,* 702,2165-2171.

Brotman, D. J., & Girod, J. P. (2002). The metabolic syndrome: A tug-of-war with no winner. *Cleveland Clinic Journal ofMedicine,* 69(12), 990-994.

Brownell, K. D., & Rodin, J. (1994). The dieting maelstrom: Is it possible and advisable to lose weight? *American Psychologist, 49(9),* 781.

Bryan, A., Hutchison, K. E., Seals, D. R., & Allen, D. L. (2007). A transdisciplinary model integrating genetic, physiological, and psychological correlates of voluntary exercise. *Health Psychology: Official Journal of the Division of Health Psychology, American Psychological Association,* 26(1), 30.

Bryant, E., Caudwell, P., Hopkins, M. E., King, N. A., & Blundell, J. E. (2012). Psycho-markers of weight loss. the roles of TFEQ disinhibition and restraint in exercise-induced weight management. *Appetite, 58*(1), 234-241.

Bryant, E., King, N., & Blundell, J. (2007). Disinhibition: Its effects on appetite and weight regulation. *Obesity Reviews, 9*, 409-419.

Buffenstein, R., Poppitt, S., McDevitt, R., & Prentice, A. (1995). Food intake and the menstrual cycle: A retrospective analysis, with implications for appetite research. *Physiology & Behavior, 58*(6), 1067-1077.

Burkman, R., Bell, C., & Serfaty, D. (2011). The evolution of combined oral contraception: Improving the risk-to-benefit ratio. *Contraception, 84*(1), 19-34.

Burton, P., Smit, H. J., & Lightowler, H. J. (2007). The influence of restrained and external eating patterns on overeating. *Appetite, 49*(1), 191-197.

Butler, J. E., Larsen, T. S., Gandevia, S. C., & Petersen, N. T. (2007). The nature of corticospinal paths driving human motoneurones during voluntary contractions. *The Journal of Physiology, 584*(2), 651-659.

Butte, N. F., Ekelund, U., & Westerterp, K. R. (2012). Assessing physical activity using wearable monitors: Measures of physical activity. *Medicine & Science in Sports & Exercise, 44*(1 suppl), S5-12.

Byrne, N. M., Meerkin, J. D., Laukkanen, R., Ross, R., Fogelholm, M., & Hills, A. P. (2006). Weight loss strategies for obese adults: Personalized weight management program vs. standard care. *Obesity, 14*(10), 1777-1788.

Caballero, B. (2007). The global epidemic of obesity: An overview. *Epidemiologic Reviews, 29*(1), 1-5.

Cappelleri, J., Bushmakin, A., Gerber, R., Leidy, N., Sexton, C., Lowe, M., et al. (2009). Psychometric analysis of the three-factor eating questionnaire-R21: Results

from a large diverse sample of obese and non-obese participants. *International Journal of Obesity, 33*(6), 611-620.

Carey, M., Markham, C., Gaffney, P., Boran, G., & Maher, V. (2006). Validation of a point of care lipid analyser using a hospital based reference laboratory. *Irish Journal of Medical Science, 175*(4), 30-35.

Caspersen, C. J., Powell, K. E., & Christenson, G. M. (1985). Physical activity, exercise, and physical fitness: Definitions and distinctions for health-related research. *Public Health Reports, 100*(2), 126.

Catenacci, V. A., & Wyatt, H. R. (2007). The role of physical activity in producing and maintaining weight loss. *Nature Clinical Practice Endocrinology & Metabolism, 3*(7), 518-529.

Caudwell, P., Finlayson, G., Gibbons, C., Hopkins, M., King, N., Näslund, E., et al. (2013a). Resting metabolic rate is associated with hunger, self-determined meal size, and daily energy intake and may represent a marker for appetite. *The American Journal of Clinical Nutrition, 97*(1), 7-14.

Caudwell, P., Gibbons, C., Hopkins, M., King, N., Finlayson, G., & Blundell, J. (2013b). No sex difference in body fat in response to supervised and measured exercise. *Medicine & Science in Sports & Exercise, 45*(2), 351-358.

Caudwell, P., Hopkins, M., King, N. A., Stubbs, R. J., & Blundell, J. E. (2009). Exercise alone is not enough: Weight loss also needs a healthy (mediterranean) diet? *Public Health Nutrition, 12*(9), 1663.

Chaput, J. P., Klingenberg, L., Rosenkilde, M., Gilbert, J. A., Tremblay, A., & Sjödin, A. (2010). Physical activity plays an important role in body weight regulation. *Journal of Obesity, 2011*

Cheer, J. F., Wassum, K. M., Heien, M. L., Phillips, P. E., & Wightman, R. M. (2004). Cannabinoids enhance subsecond dopamine release in the nucleus accumbens of awake rats. *The Journal of Neuroscience,* 24(18), 4393-4400.

Church, T. S., Martin, C. K., Thompson, A. M., Earnest, C. P., Mikus, C. R., & Blair, S. N. (2009). Changes in weight, waist circumference and compensatory responses with different doses of exercise among sedentary, overweight postmenopausal women. *PLoS One, 4(2),* e4515.

Cicchetti, D. V. (2001). Methodological commentary the precision of reliability and validity estimates re-visited: Distinguishing between clinical and statistical significance of sample size requirements. *Journal of Clinical and Experimental Neuropsychology,* 22(5), 695-700.

Colley, R. C., Hills, A. P., King, N. A., & Byrne, N. M. (2010). Exercise-induced energy expenditure: Implications for exercise prescription and obesity. *Patient Education and Counseling,* 79(3), 327-332.

Colley, R. C., Hills, A. P., O'Moore-Sullivan, T., Hickman, I. J., Prins, J. B., & Byrne, N. M. (2008). Variability in adherence to an unsupervised exercise prescription in obese women. *International Journal of Obesity, 32(5),* 837-844.

Cornier, M. A., Melanson, E. L., Salzberg, A. K., Bechtell, J. L., & Tregellas, J. R. (2012). The effects of exercise on the neuronal response to food cues. *Physiology & Behavior, 105(4),* 1028-1034.

Crouter, S., Churilla, J., & Bassett, D. (2008). Accuracy of the actiheart for the assessment of energy expenditure in adults. *European Journal of Clinical Nutrition, 62(6),* 704-711.

Cummings, D. E., Purnell, J. Q., Frayo, R. S., Schmidova, K., Wisse, B. E., & Weigle, D. S. (2001). A preprandial rise in plasma ghrelin levels suggests a role in meal initiation in humans. *Diabetes, 50*(8), 1714-1719.

Cunha, F. A., Farinatti P. T., & Midgley, A. W. (2011). Methodological and practical application issues in exercise prescription using the heart rate reserve and oxygen uptake reserve methods. *Journal of Science and Medicine in Sport / Sports Medicine Australia, 14*(1), 46-57.

D'Alessio, D., Kavle, E., Mozzoli, M., Smalley, K., Polansky, M., Kendrick, Z., et al. (1988). Thermic effect of food in lean and obese men. *Journal of Clinical Investigation, 81*(6), 1781.

Darmon, N., & Drewnowski, A. (2008). Does social class predict diet quality? *The American Journal of Clinical Nutrition, 87*(5), 1107-1117.

Darmon, N., Ferguson, E., & Briend, A. (2003). Do economic constraints encourage the selection of energy dense diets? *Appetite, 41*(3), 315-322.

Davis, J. D., & Wirtshafter, D. (1978). Set points or settling points for body weight?: A reply to mrosovsky and powley. *Behavioral Biology, 24*(3), 405-411.

Davy, B. M., Van Walleghen, E. L., & Orr, J. S. (2007). Sex differences in acute energy intake regulation. *Appetite, 49*(1), 141-147.

De Castro, J. M. (1995). The relationship of cognitive restraint to the spontaneous food and fluid intake of free-living humans. *Physiology & Behavior, 57*(2), 287-295.

De Castro, J. M. (1997). How can energy balance be achieved by free-living human subjects? *Proceedings of the Nutrition Society, 56*(1A), 1-14.

De Castro, J. M. (2010). The control of food intake of free-living humans: Putting the pieces back together. *Physiology & Behavior, 100*(5), 446.

De Castro, J. M., & Brewer, E. M. (1992). The amount eaten in meals by humans is a power function of the number of people present. *Physiology & Behavior, 51*(1), 121-125.

De Lauzon, B., Romon, M., Deschamps, V., Lafay, L., Borys, J., Karlsson, J., et al. (2004). The three-factor eating questionnaire-R18 is able to distinguish among different eating patterns in a general population. *The Journal of Nutrition, 134*(9), 2372-2380.

De Souza, R. J., Bray, G. A., Carey, V. J., Hall, K. D., LeBoff, M. S., Loria, C. M., et al. (2012). Effects of 4 weight-loss diets differing in fat, protein, and carbohydrate on fat mass, lean mass, visceral adipose tissue, and hepatic fat: Results from the POUNDS LOST trial. *The American Journal of Clinical Nutrition, 95*(3), 614-625.

Deci, E. L., & Ryan, R. M. (1985). Intrinsic motivation and self-determination in human behavior. *New York and London: Plenum Press.*

Deighton, K., Barry, R., Connon, C. E., & Stensel, D. J. (2012). Appetite, gut hormone and energy intake responses to low volume sprint interval and traditional endurance exercise. *European Journal of Applied Physiology,* , 1-10.

Deslandes, A., Moraes, H., Ferreira, C., Veiga, H., Silveira, H., Mouta, R., et al. (2009). Exercise and mental health: Many reasons to move. *Neuropsychobiology, 59*(4), 191-198.

Di Marzo, V., & Matias, I. (2005). Endocannabinoid control of food intake and energy balance. *Nature Neuroscience, 8*(5), 585-589.

Dishman, R. K., Berthoud, H., Booth, F. W., Cotman, C. W., Edgerton, V. R., Fleshner, M. R., et al. (2006). Neurobiology of exercise. *Obesity, 14*(3), 345-356.

Dishman, R. K., Sallis, J. F., & Orenstein, D. R. (1985). The determinants of physical activity and exercise. *Public Health Reports, 100*(2), 158.

Donnelly, J. E., & Smith, B. K. (2005). Is exercise effective for weight loss with ad libitum diet? energy balance, compensation, and gender differences. *Exercise and Sport Sciences Reviews, 33*(4), 169-174.

Donnelly, J. E., Jacobsen, D. J., Heelan, K. S., Seip, R., & Smith, S. (2000). The effects of 18 months of intermittent vs. continuous exercise on aerobic capacity, body weight and composition, and metabolic fitness in previously sedentary, moderately obese females. *International Journal of Obesity and Related Metabolic Disorders: Journal of the International Association for the Study of Obesity, 24*(5), 566-572.

Donnelly, J. E., Kirk, E. P., Jacobsen, D. J., Hill, J. O., Sullivan, D. K., & Johnson, S. L. (2003). Effects of 16 mo of verified, supervised aerobic exercise on macronutrient intake in overweight men and women: The midwest exercise trial. *American Journal of Clinical Nutrition, 78*(5), 950.

Downing, S. M. (2004). Reliability: On the reproducibility of assessment data. *Medical Education, 38*(9), 1006-1012.

Drenowatz, C., Eisenmann, J. C., Carlson, J. J., Pfeiffer, K. A., & Pivarnik, J. M. (2012). Energy expenditure and dietary intake during high-volume and low-volume training periods among male endurance athletes. *Applied Physiology, Nutrition, and Metabolism, 37*(2), 199-205.

Drent, M. L., Larsson, I., William-Olsson, T., Quaade, F., Czubayko, F., Von Bergmann, K., ... & Van der Veen, E. A. (1995). Orlistat (Ro 18-0647), a lipase inhibitor, in the treatment of human obesity: a multiple dose study. *International journal of obesity and related metabolic disorders: journal of the International Association for the Study of Obesity, 19*(4), 221-226.

Durnin, J. (1961). Appetite'and the relationships between expenditure and intake of calories in man. *The Journal of Physiology, 156*(2), 294-306.

Durnin, J., & Womersley, J. (1974). Body fat assessed from total body density and its estimation from skinfold thickness: Measurements on 481 men and women aged from 16 to 72 years. *British Journal of Nutrition, 32*(01), 77-97.

Dye, L., & Blundell, J. (1997). Menstrual cycle and appetite control: Implications for weight regulation. *Human Reproduction, 12*(6), 1142.

Ebbeling, C. B., Swain, J. F., Feldman, H. A., Wong, W. W., Hachey, D. L., Garcia-Lago, E., et al. (2012). Effects of dietary composition during weight loss maintenance: A controlled feeding study. *JAMA: The Journal of the American Medical Association, 307*(24), 2627.

Eck, L., Bennett, A., Egan, B., Ray, J., Mitchell, C., Smith, M., et al. (1997). Differences in macronutrient selections in users and nonusers of an oral contraceptive. *American Journal of Clinical Nutrition, 65*(2), 419.

Eckel, R. H. (2008a). Obesity research in the next decade. *International Journal of Obesity, 32*, S143-S151.

Eckel, R. H. (2008b). Nonsurgical management of obesity in adults. *New England Journal of Medicine, 358*(18), 1941-1950.

Edholm, O. (1973). Dietary data and estimates of energy expenditure. *Proceedings of the Royal Society of Medicine, 66*(7), 641.

Edholm, O., Adam, J., Healy, M., Wolff, H., Goldsmith, R., & Best, T. (1970). Food intake and energy expenditure of army recruits. *British Journal of Nutrition, 24*(04), 1091-1107.

Edholm, O., Fletcher, J., Widdowson, E. M., & McCance, R. (1955). The energy expenditure and food intake of individual men. *British Journal of Nutrition, 9*(03), 286-300.

Ekkekakis, P., Hall, E. E., & Petruzzello, S. J. (2005). Variation and homogeneity in affective responses to physical activity of varying intensities: An alternative perspective on dose–response based on evolutionary considerations. *Journal of sports sciences, 23*(5), 477-500.

Elder, S. J., & Roberts, S. B. (2007). The effects of exercise on food intake and body fatness: A summary of published studies. *Nutrition Reviews, 65*(1), 1-19.

Epstein, L. H., Dearing, K. K., Roba, L. G., & Finkelstein, E. (2010). The influence of taxes and subsidies on energy purchased in an experimental purchasing study. *Psychological Science, 21*(3), 406-414.

Figlewicz, D. P., & Benoit, S. C. (2009). Insulin, leptin, and food reward: Update 2008. *American Journal of Physiology-Regulatory, Integrative and Comparative Physiology, 296*(1), R9-R19.

Finlayson, G., Bryant, E., Blundell, J. E., & King, N. A. (2009). Acute compensatory eating following exercise is associated with implicit hedonic wanting for food. *Physiology & Behavior, 97*(1), 62-67.

Finucane, M. M., Stevens, G. A., Cowan, M. J., Danaei, G., Lin, J. K., Paciorek, C. J., et al. (2011). National, regional, and global trends in body-mass index since 1980: Systematic analysis of health examination surveys and epidemiological studies with 960 country-years and 9· 1 million participants. *The Lancet, 377*(9765), 557-567.

Flint, A., Raben, A., Blundell, J. E., & Astrup, A. (2000). Reproducibility, power and validity of visual analogue scales in assessment of appetite sensations in single test meal studies. *International Journal of Obesity and Related Metabolic Disorders: Journal of the International Association for the Study of Obesity, 24*(1), 38-48.

Fogelholm, M., & Kukkonen-Harjula, K. (2000). Does physical activity prevent weight gain–a systematic review. *Obesity reviews, 1*(2), 95-111

Foley, L., Maddison, R., Olds, T., & Ridley, K. (2012). Self-report use-of-time tools for the assessment of physical activity and sedentary behaviour in young people: Systematic review. *Obesity Reviews, 13*(8), 711-722.

Foster, G. D., Makris, A. P., & Bailer, B. A. (2005). Behavioral treatment of obesity. *The American Journal of Clinical Nutrition, 82*(1), 230S-235S.

Foster, G. D., Wadden, T. A., Peterson, F. J., Letizia, K. A., Bartlett, S. J., & Conill, A. M. (1992). A controlled comparison of three very-low-calorie diets: Effects on weight, body composition, and symptoms. *The American Journal of Clinical Nutrition, 55*(4), 811-817.

Foster, G., Wadden, T., Swain, R., Stunkard, A., Platte, P., & Vogt, R. (1998). The eating inventory in obese women: Clinical correlates and relationship to weight loss. *International Journal of Obesity, 22*(8), 778-785.

Foster-Schubert, K. E., McTiernan, A., Frayo, R. S., Schwartz, R. S., Rajan, K. B., Yasui, Y., et al. (2005). Human plasma ghrelin levels increase during a one-year exercise program. *Journal of Clinical Endocrinology & Metabolism, 90*(2), 820-825.

Fox, K. R. (1999). The influence of physical activity on mental well-being. *Public Health Nutrition, 2*(3a), 411-418.

Frank, L. D., Schmid, T. L., Sallis, J. F., Chapman, J., & Saelens, B. E. (2005). Linking objectively measured physical activity with objectively measured urban form: Findings from SMARTRAQ. *American Journal of Preventive Medicine, 28*(2), 117-125.

Franz, M., Vanwormer, J., Crain, L., Boucher, J., Histon, T., Capland, W., et al. (2007). Weight-loss outcomes: A systematic review and meta-analysis of weight-loss clinical trials with a minimum 1-year follow-up. *Journal of the American Dietetic Association, 107*, 1755-1767.

Frayn, K. N. (1983). Calculation of substrate oxidation rates in vivo from gaseous exchange. *Journal of Applied Physiology, 55*(2), 628-634.

Frey-Hewitt, B., Vranizan, K., Dreon, D., & Wood, P. (1990). The effect of weight loss by dieting or exercise on resting metabolic rate in overweight men. *International Journal of Obesity, 14*(4), 327.

Fried, M., Hainer, V., Basdevant, A., Buchwald, H., Deitel, M., Finer, N., et al. (2007). Interdisciplinary european guidelines for surgery for severe (morbid) obesity. *Obesity Surgery, 17*(2), 260-270.

Fulton, S. (2010). Appetite and reward. *Frontiers in Neuroendocrinology, 31*(1), 85-103.

Galgani, J., & Ravussin, E. (2008). Energy metabolism, fuel selection and body weight regulation. *International Journal of Obesity, 32*, S109-S119.

Gallo, M., Lopez, L., Grimes, D., Schulz, K., & Helmerhorst, F. (2011). Combination contraceptives: Effects on weight (review). *Cochrane Database of Systematic Reviews,* Issue 9. Art. No.: CD003987. DOI: 10.1002/14651858.CD003987.pub4

Gardner, B., Bruijn, G. d., & Lally, P. (2011). A systematic review and meta-analysis of applications of the self-report habit index to nutrition and physical activity behaviours. *Annals of Behavioral Medicine, 42*(2), 174-187.

Garfield, A. S., Lam, D. D., Marston, O. J., Przydzial, M. J., & Heisler, L. K. (2009). Role of central melanocortin pathways in energy homeostasis. *Trends in Endocrinology & Metabolism, 20*(5), 203-215.

Garland, T., Schutz, H., Chappell, M. A., Keeney, B. K., Meek, T. H., Copes, L. E., et al. (2011). The biological control of voluntary exercise, spontaneous physical activity and daily energy expenditure in relation to obesity: Human and rodent perspectives. *The Journal of Experimental Biology, 214*(2), 206-229.

Geliebter, A., & Aversa, A. (2003). Emotional eating in overweight, normal weight, and underweight individuals. *Eating Behaviors, 3*(4), 341-347.

George, V. A., & Morganstein, A. (2003). Effect of moderate intensity exercise on acute energy intake in normal and overweight females. *Appetite, 40*(1), 43-46.

Gilhooly, C., Das, S., Golden, J., McCrory, M., Dallal, G., Saltzman, E., et al. (2007). Food cravings and energy regulation: The characteristics of craved foods and their relationship with eating behaviors and weight change during 6 months of dietary energy restriction. *International Journal of Obesity, 31*(12), 1849-1858.

Gilsenan, M., Murgatroyd, P., Leahy, F., Goldberg, G., & Prentice, A. (1998). The response of energy intake and macronutrient balance to manipulation of physical activity levels in lean men. *Proceedings-Nutrition Society of London, 57*, 19A.

Glandt, M., & Raz, I. (2011). Present and future: Pharmacologic treatment of obesity. *Journal of Obesity, 2011*

Glass, S., & Dwyer, G. B. (Eds.). (2007). *ACSM's Metabolic Calculations Handbook.* Lippincott Williams & Wilkins.

Godin, G., & Shephard, R. J. (1985). A simple method to assess exercise behavior in the community. *Canadian Journal of Applied Sport Sciences.Journal Canadien Des Sciences Appliquees Au Sport, 10*(3), 141.

Goldberg, G., Black, A., Jebb, S., Cole, T., Murgatroyd, P., Coward, W., et al. (1991). Critical evaluation of energy intake data using fundamental principles of energy physiology: 1. derivation of cut-off limits to identify under-recording. *European Journal of Clinical Nutrition, 45*(12), 569.

Goran, M. I. (2000). Energy metabolism and obesity. *Medical Clinics of North America, 84*(2), 347-362.

Goran, M., & Poehlman, E. (1992). Endurance training does not enhance total energy expenditure in healthy elderly persons. *American Journal of Physiology- Endocrinology and Metabolism, 263*(5), E950-957.

Gore, C., Norton, K., Olds, T., Whittingham, N., Birchall, K., Clough, M., et al. (1996). Accreditation in anthropometry: An Australian model. In K. Norton, & T. Olds (Eds.), (pp. 395-411) University of New South Wales Press Sydney.

Gorin, A. A., Wing, R. R., Fava, J. L., Jakicic, J. M., Jeffery, R., West, D. S., et al. (2008). Weight loss treatment influences untreated spouses and the home environment: Evidence of a ripple effect. *International Journal of Obesity, 32*(11), 1678-1684.

Goto, R., & Mascie-Taylor, C. G. N. (2007). Precision of measurement as a component of human variation. *Journal of Physiological Anthropology, 26*(2), 253-256.

Grace, C. (2011). A review of one - to - one dietetic obesity management in adults. *Journal of Human Nutrition and Dietetics, 24*(1), 13-22.

Groesz, L. M., McCoy, S., Carl, J., Saslow, L., Stewart, J., Adler, N., et al. (2012). What is eating you? Stress and the drive to eat. *Appetite, 58*(2), 717-721.

Guelfi, K. J., Donges, C. E., & Duffield, R. (2012). Beneficial effects of 12 weeks of aerobic compared with resistance exercise training on perceived appetite in previously sedentary overweight and obese men. *Metabolism Clinical and Experimental,62,* 235-243

Hagobian, T. A., & Braun, B. (2010). Physical activity and hormonal regulation of appetite: Sex differences and weight control. *Exercise and Sport Sciences Reviews, 38*(1), 25-30.

Hagobian, T. A., Yamashiro, M., Hinkel-Lipsker, J., Streder, K., Evero, N., & Hackney, T. (2012). Effects of acute exercise on appetite hormones and ad libitum energy intake in men and women. *Applied Physiology, Nutrition, and Metabolism, 38*(999), 66-72.

Hall, K. D., Heymsfield, S. B., Kemnitz, J. W., Klein, S., Schoeller, D. A., & Speakman, J. R. (2012). Energy balance and its components: Implications for body weight regulation. *The American Journal of Clinical Nutrition, 95*(4), 989-994.

Hall, K. D., Sacks, G., Chandramohan, D., Chow, C. C., Wang, Y. C., Gortmaker, S. L., et al. (2011). Quantification of the effect of energy imbalance on bodyweight. *The Lancet, 378*(9793), 826-837.

Hamani, Y., Sciaki-Tamir, Y., Deri-Hasid, R., Miller-Pogrund, T., Milwidsky, A., & Haimov-Kochman, R. (2007). Misconceptions about oral contraception pills among adolescents and physicians. *Human Reproduction, 22*(12), 3078-3083.

Han, T., Van Leer, E., Seidell, J., & Lean, M. (1995). Waist circumference action levels in the identification of cardiovascular risk factors: Prevalence study in a random sample. *British Medical Journal, 311*(7017), 1401-1405.

Harris, C. L., & George, V. A. (2008). The impact of dietary restraint and moderate-intensity exercise on post-exercise energy intake in sedentary males. *Eating Behaviors, 9*(4), 415-422.

Harris, R. (1990). Role of set-point theory in regulation of body weight. *The Journal of the Federation of American Societies for Experimental Biology, 4*(15), 3310-3318.

Harvey, J., Wing, R. R., & Mullen, M. (1993). Effects on food cravings of a very low calorie diet or a balanced, low calorie diet. *Appetite, 21*(2), 105-115.

Haskell, W. L., Blair, S. N., & Hill, J. O. (2009). Physical activity: health outcomes and importance for public health policy. *Preventive medicine, 49*(4), 280-282.

Haslam, D. (2007). Obesity: A medical history. *Obesity Reviews, 8*, 31-36.

Haslam, D. W., & James, W. P. T. (2005). Life expectancy. *Lancet, 366*, 1197-1209.

Haug, E., Torsheim, T., Sallis, J. F., & Samdal, O. (2010). The characteristics of the outdoor school environment associated with physical activity. *Health Education Research, 25*(2), 248-256.

Hensrud, D. D. (2012). Dietary treatment and Long - Term weight loss and maintenance in type 2 diabetes. *Obesity Research, 9*(S4), 348S-353S.

Herman, C. P., & Polivy, J. (2005). Normative influences on food intake. *Physiology & Behavior, 86*(5), 762-772.

Hetherington, M. M. (2007). Cues to overeat: Psychological factors influencing overconsumption. *Proceedings of the Nutrition Society, 66*(01), 113-123.

Hetherington, M. M., Foster, R., Newman, T., Anderson, A. S., & Norton, G. (2006). Understanding variety: Tasting different foods delays satiation. *Physiology & Behavior, 87*(2), 273-271.

Heymsfield, S. B., McManus, C., Stevens, V., & Smith, J. (1982). Muscle mass: Reliable indicator of protein-energy malnutrition severity and outcome. *The American Journal of Clinical Nutrition, 35*(5), 1192-1199.

Heymsfield, S., Van Mierlo, C., Van Der Knaap, H., Heo, M., & Frier, H. (2003). Weight management using a meal replacement strategy: Meta and pooling analysis from six studies. *International Journal of Obesity, 27*(5), 537-549.

Hiilloskorpi, H., Fogelholm, M., Laukkanen, R., Pasanen, M., Oja, P., Mänttäri, A., et al. (1999). Factors affecting the relation between heart rate and energy expenditure during exercise. *International Journal of Sports Medicine, 20*(07), 438-443.

Hill, A. J., Weaver, C. F., & Blundell, J. E. (1991). Food craving, dietary restraint and mood. *Appetite, 17*(3), 187-197.

Hill, J. O., & Peters, J. C. (1998). Environmental contributions to the obesity epidemic. *Science, 280*(5368), 1371-1374.

Hill, J. O., & Wyatt, H. R. (2005). Role of physical activity in preventing and treating obesity. *Journal of Applied Physiology, 99*(2), 765-770.

Hirschberg, A. L., Byström, B., Carlström, K., & von Schoultz, B. (1996). Reduced serum cholecystokinin and increase in body fat during oral contraception. *Contraception, 53*(2), 109-113.

Hollowell, R. P., Willis, L. H., Slentz, C. A., Topping, J. D., Bhakpar, M., & Kraus, W. E. (2009). Effects of exercise training amount on physical activity energy expenditure. *Medicine & Science in Sports & Exercise, 41*(8), 1640-1644.

Hopkins, W. G. (2000). Measures of reliability in sports medicine and science. *Sports Medicine, 30*(1), 1-15.

Hosoda, H., Kojima, M., Matsuo, H., & Kangawa, K. (2000). Ghrelin and des-acyl ghrelin: Two major forms of rat ghrelin peptide in gastrointestinal tissue. *Biochemical and Biophysical Research Communications, 279*(3), 909-913.

Howley, E. T. (2001). Type of activity: resistance, aerobic and leisure versus occupational physical activity. *Medicine & Science in Sports & Exercise*, 33, S364–9.

Hubel, R., Laessle, R., Lehrke, S., & Jass, J. (2006). Laboratory measurement of cumulative food intake in humans: Results on reliability. *Appetite, 46*(1), 57-62.

Hubert, P., King, N., & Blundell, J. (1998). Uncoupling the effects of energy expenditure and energy intake: Appetite response to short-term energy deficit induced by meal omission and physical activity. *Appetite, 31*(1), 9-19.

Hunter, G. R., Wetzstein, C. J., Fields, D. A., Brown, A., & Bamman, M. M. (2000). Resistance training increases total energy expenditure and free-living physical activity in older adults. *Journal of Applied Physiology, 89*(3), 977-984.

Imbeault, P., Saint-Pierre, S., AlméRas, N., & Tremblay, A. (1997). Acute effects of exercise on energy intake and feeding behaviour. *British Journal of Nutrition, 77*(04), 511-521.

Inbar, O., Oren, A., Scheinowitz, M., Rotstein, A., Dlin, R., & Casaburi, R. (1994). Normal cardiopulmonary responses during incremental exercise in 20-to 70-yr-old men. *Medicine & Science in Sports & Exercise, 26*(5), 538-546.

Ioannides-Demos, L. L., Piccenna, L., & McNeil, J. J. (2011). Pharmacotherapies for obesity: Past, current, and future therapies. *Journal of Obesity,* 2011:179674

Irvine, M. A., Brunstrom, J. M., Gee, P., & Rogers, P. J. (2012). Increased familiarity with eating a food to fullness underlies increased expected satiety. *Appetite, 61*(1), 13-18.

Irwin, M. L., Yasui, Y., Ulrich, C. M., Bowen, D., Rudolph, R. E., Schwartz, R. S., et al. (2003). Effect of exercise on total and intra-abdominal body fat in postmenopausal women: A randomized controlled trial. *JAMA: the Journal of the American Medical Association, 289*(3), 323-330.

Jakicic, J. M. (2002). The role of physical activity in prevention and treatment of body weight gain in adults. *The Journal of Nutrition, 132*(12), 3826S-3829S.

Jakicic, J. M., & Otto, A. D. (2006). Treatment and prevention of obesity: What is the role of exercise? *Nutrition Reviews, 64*, S57-S61.

Jakicic, J. M., Marcus, B. H., Gallagher, K. I., Napolitano, M., & Lang, W. (2003). Effect of exercise duration and intensity on weight loss in overweight, sedentary women: A randomized trial. *JAMA: the Journal of the American Medical Association, 290*(10), 1323-1330.

Jéquier, E. (2002). Leptin signaling, adiposity, and energy balance. *Annals of the New York Academy of Sciences, 967*(1), 379-388.

Jéquier, E., & Tappy, L. (1999). Regulation of body weight in humans. *Physiological Reviews, 79*(2), 451-480.

Johnson, R. K. (2002). Dietary intake—how do we measure what people are really eating? *Obesity Research, 10*(S11), 63S-68S.

Johnson, R. K. (2002). Dietary intake—how do we measure what people are really eating? *Obesity Research, 10*(S11), 63S-68S.

Johnson, W. G., Carr-Nangle, R. E., & Bergeron, K. C. (1995). Macronutrient intake, eating habits, and exercise as moderators of menstrual distress in healthy women. *Psychosomatic Medicine, 57*(4), 324-330.

Jokisch, E., Coletta, A., & Raynor, H. A. (2012). Acute energy compensation and macronutrient intake following exercise in active and inactive males who are normal weight. *Appetite, 58*(2), 722-729.

Jonge, X. A. K. J. (2003). Effects of the menstrual cycle on exercise performance. *Sports Medicine, 33*(11), 833-851.

Joosen, A. M., Gielen, M., Vlietinck, R., & Westerterp, K. R. (2005). Genetic analysis of physical activity in twins. *The American Journal of Clinical Nutrition, 82*(6), 1253-1259.

Jorgensen, T., Andersen, L. B., Froberg, K., Maeder, U., von Huth Smith, L., & Aadahl, M. (2009). Position statement: Testing physical condition in a population–how good are the methods? *European Journal of Sport Science, 9*(5), 257-267.

Jumpertz, R., Le, D. S., Turnbaugh, P. J., Trinidad, C., Bogardus, C., Gordon, J. I., et al. (2011). Energy-balance studies reveal associations between gut microbes, caloric load, and nutrient absorption in humans. *The American Journal of Clinical Nutrition, 94*(1), 58-65.

Kang, J. G., & Park, C. Y. (2012). Anti-obesity drugs: A review about their effects and safety. *Diabetes & Metabolism Journal, 36*(1), 13-25.

Karlsson, J., Persson, L. O., Sjostrom, L., & Sullivan, M. (2000). Psychometric properties and factor structure of the three-factor eating questionnaire (TFEQ) in obese men and women. results from the swedish obese subjects (SOS) study. *International Journal of Obesity and Related Metabolic Disorders: Journal of the International Association for the Study of Obesity, 24*(12), 1715-1725.

Karlsson, R., Lindén, A., & von Schoultz, B. (1992). Suppression of 24-hour cholecystokinin secretion by oral contraceptives. *American Journal of Obstetrics and Gynecology, 167*(1), 58-59.

Katzmarzyk, P. T., Janssen, I., Ross, R., Church, T. S., & Blair, S. N. (2006). The importance of waist circumference in the definition of metabolic syndrome prospective analyses of mortality in men. *Diabetes Care, 29*(2), 404-409.

Kaye, W. H., Wierenga, C. E., Bailer, U. F., Simmons, A. N., & Bischoff-Grethe, A. (2013). Nothing tastes as good as skinny feels: The neurobiology of anorexia nervosa. *Trends in Neurosciences, 36*(2), 110-120.

Keim, N. L., Canty, D. J., Barbieri, T. F., & Wu, M. M. (1996). Effect of exercise and dietary restraint on energy intake of reduced-obese women. *Appetite, 26*(1), 55-70.

Keim, N., Barbieri, T., & Belko, A. (1990). The effect of exercise on energy intake and body composition in overweight women. *International Journal of Obesity, 14*(4), 335-346.

Kelly, P. J., Guelfi, K. J., Wallman, K. E., & Fairchild, T. J. (2012). Mild dehydration does not reduce post-exercise appetite or energy intake. *Medicine & Science in Sports & Exercise, 44*(3), 516–524.

Kennedy, G. C. (1953). The role of depot fat in the hypothalamic control of food intake in the rat. *Proceedings of the Royal Society of London. Series B-Biological Sciences, 140*(901), 578-592.

Kesäniemi, A., Riddoch, C. J., Reeder, B., Blair, S. N., & Sørensen, T. I. A. (2010). Advancing the future of physical activity guidelines in Canada: An independent expert panel interpretation of the evidence, *International Journal of Behavioral Nutrition and Physical Activity, 7*(1), 41.

Keytel, L. R., Lambert, M. I., Johnson, J., Noakes, T. D., & Lambert, E. V. (2001). Free living energy expenditure in post menopausal women before and after exercise training. *International Journal of Sport Nutrition and Exercise Metabolism, 11*(2), 226-237.

Keytel, L., Goedecke, J., Noakes, T., Hiiloskorpi, H., Laukkanen, R., Van Der Merwe, L., et al. (2005). Prediction of energy expenditure from heart rate monitoring during submaximal exercise. *Journal of Sports Sciences, 23*(3), 289-297.

King, J. A., Miyashita, M., Wasse, L. K., & Stensel, D. J. (2010a). Influence of prolonged treadmill running on appetite, energy intake and circulating concentrations of acylated ghrelin. *Appetite, 54*(3), 492-498.

King, J. A., Wasse, L. K., & Stensel, D. J. (2013). Acute exercise increases feeding latency in healthy normal weight young males but does not alter energy intake. *Appetite, 61*(1), 45-51.

King, J. A., Wasse, L. K., Broom, D. R., & Stensel, D. J. (2010b). Influence of brisk walking on appetite, energy intake, and plasma acylated ghrelin. *Medicine & Science in Sports & Exercise, 42*(3), 485-492.

King, J. A., Wasse, L. K., Ewens, J., Crystallis, K., Emmanuel, J., Batterham, R. L., et al. (2011). Differential acylated ghrelin, peptide YY3–36, appetite, and food intake

responses to equivalent energy deficits created by exercise and food restriction. *Journal of Clinical Endocrinology & Metabolism, 96*(4), 1114-1121.

King, N. A. (1999). What processes are involved in the appetite response to moderate increases in exercise-induced energy expenditure? *Proceedings of the Nutrition Society, 58*(01), 107-113.

King, N. A., Appleton, K., Rogers, P. J., & Blundell, J. E. (1999). Effects of sweetness and energy in drinks on food intake following exercise. *Physiology & Behavior, 66*(2), 375-379.

King, N. A., Caudwell, P. P., Hopkins, M., Stubbs, J. R., Naslund, E., & Blundell, J. E. (2009). Dual-process action of exercise on appetite control: Increase in orexigenic drive but improvement in meal-induced satiety. *The American Journal of Clinical Nutrition, 90*(4), 921-927.

King, N. A., Caudwell, P., Hopkins, M., Byrne, N. M., Colley, R., Hills, A. P., et al. (2007). Metabolic and behavioral compensatory responses to exercise interventions: Barriers to weight loss. *Obesity, 15*(6), 1373-1383.

King, N. A., Hopkins, M., Caudwell, P., Stubbs, R., & Blundell, J. E. (2008). Individual variability following 12 weeks of supervised exercise: Identification and characterization of compensation for exercise-induced weight loss. *International Journal of Obesity, 32*(1), 177-184.

King, N. A., Snell, L., Smith, R. D., & Blundell, J. E. (1996). Effects of short-term exercise on appetite responses in unrestrained females. *European Journal of Clinical Nutrition, 50*(10), 663-667.

King, N., & Blundell, J. (1995). High-fat foods overcome the energy expenditure induced by high-intensity cycling or running. *European Journal of Clinical Nutrition, 49*(2), 114.

King, N., Burley, V., & Blundell, J. (1994). Exercise-induced suppression of appetite: Effects on food intake and implications for energy balance. *European Journal of Clinical Nutrition, 48*(10), 715-724.

King, N., Horner, K., Hills, A., Byrne, N., Wood, R., Bryant, E., et al. (2012). Exercise, appetite and weight management: Understanding the compensatory responses in eating behaviour and how they contribute to variability in exercise-induced weight loss. *British Journal of Sports Medicine, 46*(5), 315-322.

King, N., Lluch, A., Stubbs, R., & Blundell, J. (1997). High dose exercise does not increase hunger or energy intake in free living males. *European Journal of Clinical Nutrition, 51*(7), 478-483.

Kirkpatrick, S., & Tarasuk, V. (2003). The relationship between low income and household food expenditure patterns in Canada. *Public Health Nutrition, 6*(6), 589-598.

Kissileff, H., Klingsberg, G., & Van Itallie, T. (1980). Universal eating monitor for continuous recording of solid or liquid consumption in man. *American Journal of Physiology- Regulatory, Integrative and Comparative Physiology, 238*(1), R14-R22.

Kissileff, H., Pi-Sunyer, F., Segal, K., Meltzer, S., & Foelsch, P. (1990). Acute effects of exercise on food intake in obese and nonobese women. *American Journal of Clinical Nutrition, 52*(2), 240-245.

Klausen, B., Toubro, S., Ranneries, C., Rehfeld, J., Holst, J., Christensen, N., et al. (1999). Increased intensity of a single exercise bout stimulates subsequent fat intake. *International Journal of Obesity, 23*(12), 1282-1287.

Klein, S., Allison, D. B., Heymsfield, S. B., Kelley, D. E., Leibel, R. L., Nonas, C., et al. (2007). Waist circumference and cardiometabolic risk: A consensus statement from shaping America's health: Association for Weight Management and Obesity Prevention;

NAASO, the Obesity Society; the American Society for Nutrition; and the American Diabetes Association. *Obesity, 15*(5), 1061-1067.

Knittle, J., Timmers, K., Ginsberg-Fellner, F., Brown, R., & Katz, D. (1979). The growth of adipose tissue in children and adolescents. cross-sectional and longitudinal studies of adipose cell number and size. *Journal of Clinical Investigation, 63*(2), 239.

Koeneman, M. A., Verheijden, M. W., Chinapaw, M. J., & Hopman-Rock, M. (2011). Determinants of physical activity and exercise in healthy older adults: A systematic review. *International Journal of Behavioral Nutrition and Physical Activity, 8*(1), 142.

Könner, A. C., Klöckener, T., & Brüning, J. C. (2009). Control of energy homeostasis by insulin and leptin: Targeting the arcuate nucleus and beyond. *Physiology & Behavior, 97*(5), 632-638.

Kotz, C. M., Teske, J. A., & Billington, C. J. (2008). Neuroregulation of nonexercise activity thermogenesis and obesity resistance. *American Journal of Physiology-Regulatory, Integrative and Comparative Physiology, 294*(3), R699-R710.

Koulouri, A., Tigbe, W., & Lean, M. (2006). The effect of advice to walk 2000 extra steps daily on food intake. *Journal of Human Nutrition and Dietetics, 19*(4), 263-266.

Kremers, S. P., De Bruijn, G., Visscher, T. L., Van Mechelen, W., De Vries, N. K., & Brug, J. (2006). Environmental influences on energy balance-related behaviors: A dual-process view. *International Journal of Behavioral Nutrition and Physical Activity, 3*(1), 9.

Kubik, M. Y., Lytle, L. A., Hannan, P. J., Perry, C. L., & Story, M. (2003). The association of the school food environment with dietary behaviors of young adolescents. *Journal Information, 93*(7)

Larson-Meyer, D. E., Palm, S., Bansal, A., Austin, K. J., Hart, A. M., & Alexander, B. M. (2012). Influence of running and walking on hormonal regulators of appetite in women. *Journal of Obesity,*

Lauderdale, D. S., Fabsitz, R., Meyer, J. M., Sholinsky, P., Ramakrishnan, V., & Goldberg, J. (1997). Familial determinants of moderate and intense physical activity: A twin study. *Medicine & Science in Sports & Exercise, 29*(8), 1062.

Lawson, O. J., Williamson, D. A., Champagne, C. M., DeLany, J. P., Brooks, E. R., Howat, P. M., et al. (1995). The association of body weight, dietary intake, and energy expenditure with dietary restraint and disinhibition. *Obesity Research, 3*(2), 153-161.

Leamy, L. J., Pomp, D., & Lightfoot, J. T. (2008). An epistatic genetic basis for physical activity traits in mice. *Journal of Heredity, 99*(6), 639-646.

Leddy, J. J., Epstein, L. H., Jaroni, J. L., Roemmich, J. N., Paluch, R. A., Goldfield, G. S., et al. (2004). Influence of methylphenidate on eating in obese men. *Obesity Research, 12*(2), 224-232.

Leon, A. S., Conrad, J., Hunninghake, D., & Serfass, R. (1979). Effects of a vigorous walking program on body composition, and carbohydrate and lipid metabolism of obese young men. *The American Journal of Clinical Nutrition, 32*(9), 1776-1787.

Levine, J. A. (2003). Non-exercise activity thermogenesis. *Proceedings of the Nutrition Society, 62*(03), 667-679.

Levine, J. A. (2004). Nonexercise activity thermogenesis (NEAT): Environment and biology. *American Journal of Physiology- Endocrinology and Metabolism, 286*(5), E675-E685.

Levine, J. A. (2005). Measurement of energy expenditure. *Public Health Nutrition, 8*(7a), 1123-1132.

Levine, J. A., Eberhardt, N. L., & Jensen, M. D. (1999). Role of nonexercise activity thermogenesis in resistance to fat gain in humans. *Science, 283*(5399), 212-214.

Levine, J. A., Schleusner, S. J., & Jensen, M. D. (2000). Energy expenditure of nonexercise activity. *American Journal of Clinical Nutrition, 72*(6), 1451-1454.

Levine, J. A., Vander Weg, M. W., Hill, J. O., & Klesges, R. C. (2006). Non-exercise activity thermogenesis the crouching tiger hidden dragon of societal weight gain. *Arteriosclerosis, Thrombosis, and Vascular Biology, 26*(4), 729-736.

Levine, J., & Kotz, C. (2005). NEAT-non-exercise activity thermogenesis-egocentric &geocentric environmental factors vs. biological regulation. *Acta Physiologica Scandinavica, 184*(4), 309-318.

Levitsky, D. A. (2002). Putting behavior back into feeding behavior: A tribute to george collier. *Appetite, 38*(2), 143-148.

Lightfoot, J. T. (2008). Sex hormones' regulation of rodent physical activity: A review. *International Journal of Biological Sciences, 4*(3), 126-132.

Lightfoot, J. T., Turner, M. J., Daves, M., Vordermark, A., & Kleeberger, S. R. (2004). Genetic influence on daily wheel running activity level. *Physiological Genomics, 19*(3), 270-276.

Lightfoot, J. T., Turner, M. J., Pomp, D., Kleeberger, S. R., & Leamy, L. J. (2008). Quantitative trait loci for physical activity traits in mice. *Physiological Genomics, 32*(3), 401-408.

Livingstone, M. B. E., & Black, A. E. (2003). Markers of the validity of reported energy intake. *The Journal of Nutrition, 133*(3), 895S-920S.

Livingstone, M., Strain, J., Prentice, A., Coward, W., Nevin, G., Barker, M., et al. (1991). Potential contribution of leisure activity to the energy expenditure patterns of sedentary populations. *British Journal of Nutrition, 65*, 145-155.

Lluch, A., King, N. A., & Blundell, J. E. (2000). No energy compensation at the meal following exercise indietary restrained and unrestrained women. *British Journal of Nutrition, 84*(02), 219-225.

Lluch, A., King, N., & Blundell, J. (1998). Exercise in dietary restrained women: No effect on energy intake but change in hedonic ratings. *European Journal of Clinical Nutrition, 52*(4), 300-307.

Long, S., Hart, K., & Morgan, L. (2002). The ability of habitual exercise to influence appetite and food intake in response to high-and low-energy preloads in man. *British Journal of Nutrition, 87*(05), 517-523.

Lovasi, G. S., Hutson, M. A., Guerra, M., & Neckerman, K. M. (2009). Built environments and obesity in disadvantaged populations. *Epidemiologic Reviews, 31*(1), 7-20.

Lowe, M. R., van Steenburgh, J., Ochner, C., & Coletta, M. (2009). Neural correlates of individual differences related to appetite. *Physiology & Behavior, 97*(5), 561-571.

Lutter, M., & Nestler, E. J. (2009). Homeostatic and hedonic signals interact in the regulation of food intake. *The Journal of Nutrition, 139*(3), 629-632.

Macdiarmid, J., & Blundell, J. (1998). Assessing dietary intake: Who, what and why of under-reporting. *Nutrition Research Reviews, 11*(2), 231-254.

Macsween, A. (2001). The reliability and validity of the astrand nomogram and linear extrapolation for deriving VO2max from submaximal exercise data. *The Journal of Sports Medicine and Physical Fitness, 41*(3), 312.

Magni, P., Dozio, E., Ruscica, M., Celotti, F., Masini, M. A., Prato, P., et al. (2009). Feeding behavior in mammals including humans. *Annals of the New York Academy of Sciences, 1163*(1), 221-232.

Mann, T., & Ward, A. (2004). To eat or not to eat: Implications of the attentional myopia model for restrained eaters. *Journal of Abnormal Psychology, 113*(1), 90-98.

Mann, T., & Ward, A. (2007). Attention, self-control, and health behaviors. *Current Directions in Psychological Science, 16*(5), 280-283.

Manthou, E., Gill, J. M., Wright, A., & Malkova, D. (2010). Behavioural compensatory adjustments to exercise training in overweight women. *Medicine & Science in Sports & Exercise, 42*(6), 1121-1128.

Maraki, M., Tsofliou, F., Pitsiladis, Y., Malkova, D., Mutrie, N., & Higgins, S. (2005). Acute effects of a single exercise class on appetite, energy intake and mood. Is there a time of day effect? *Appetite, 45*(3), 272-278.

Marfell-Jones, M., Olds, T., Stewart, A. and Carter, J. E. L. (2006) *International standards for anthropometric assessment.* North-West University; Potchefstroom, RSA.

Martin, C. K., O'Neil, P. M., Tollefson, G., Greenway, F. L., & White, M. A. (2008). The association between food cravings and consumption of specific foods in a laboratory taste test. *Appetite, 51*(2), 324.

Martin, C. K., O'Neil, P. M., & Pawlow, L. (2006). Changes in food cravings during Low - Calorie and Very - Low - Calorie diets. *Obesity, 14*(1), 115-121.

Martins, C., Kulseng, B., King, N., Holst, J. J., & Blundell, J. (2010). The effects of exercise-induced weight loss on appetite-related peptides and motivation to eat. *Journal of Clinical Endocrinology & Metabolism, 95*(4), 1609-1616.

Martins, C., Kulseng, B., Rehfeld, J. F., King, N. A., & Blundell, J. E. (2013). Impact of chronic exercise on appetite control in overweight and obese individuals. *Medicine & Science in Sports & Exercise, 45*(5), 805-812.

Martins, C., Morgan, L. M., Bloom, S. R., & Robertson, M. D. (2007a). Effects of exercise on gut peptides, energy intake and appetite. *Journal of Endocrinology, 193*, 251-258.

Martins, C., Morgan, L., & Truby, H. (2008). A review of the effects of exercise on appetite regulation: An obesity perspective. *International Journal of Obesity, 32*, 1337-1347.

Martins, C., Truby, H., & Morgan, L. M. (2007b). Short-term appetite control in response to a 6-week exercise programme in sedentary volunteers. *British Journal of Nutrition, 98*(04), 834-842.

Matsubara, M., Sakata, I., Wada, R., Yamazaki, M., Inoue, K., & Sakai, T. (2004). Estrogen modulates ghrelin expression in the female rat stomach. *Peptides, 25*(2), 289-297.

Maurer, J., Taren, D. L., Teixeira, P. J., Thomson, C. A., Lohman, T. G., Going, S. B., et al. (2006). The psychosocial and behavioral characteristics related to energy misreporting. *Nutrition Reviews, 64*(2), 53-66.

Mayer, J. (1955). Regulation of energy intake and the body weight: The glucostatic theory and the lipostatic hypothesis. *Annals of the New York Academy of Sciences, 63*(1), 15-43.

Mayer, J., Roy, P., & Mitra, K. P. (1956). Relation between caloric intake, body weight, and physical work: Studies in an industrial male population in west bengal. *American Journal of Clinical Nutrition, 4*(2), 169.

McLaughlin, R., Malkova, D., & Nimmo, M. (2006). Spontaneous activity responses to exercise in males and females. *European Journal of Clinical Nutrition, 60*(9), 1055-1061.

McVay, M., Copeland, A., & Geiselman, P. (2011). Eating disorder pathology and menstrual cycle fluctuations in eating variables in oral contraceptive users and non-users. *Eating Behaviors, 12*, 49-55.

Meijer, E. P., Westerterp, K. R., & Verstappen, F. T. J. (1999). Effect of exercise training on total daily physical activity in elderly humans. *European Journal of Applied Physiology and Occupational Physiology, 80*(1), 16-21.

Meijer, E., Westerterp, K., & Verstappen, F. (2000). Effect of exercise training on physical activity and substrate utilization in the elderly. *International Journal of Sports Medicine, 21*(7), 499-504.

Meijer, G. A. L., Janssen, G. M. E., Westerterp, K. R., Verhoeven, F., Saris, W. H. M., & Ten Hoor, F. (1991). The effect of a 5-month endurance-training programme on physical activity: Evidence for a sex-difference in the metabolic response to exercise. *European Journal of Applied Physiology and Occupational Physiology, 62*(1), 11-17.

Mela, D. J. (2006). Eating for pleasure or just wanting to eat? reconsidering sensory hedonic responses as a driver of obesity. *Appetite, 47*(1), 10-17.

Mellinkoff, S. M., Frankland, M., Boyle, D., & Greipel, M. (1956). Relationship between serum amino acid concentration and fluctuations in appetite. *Journal of Applied Physiology, 8*(5), 535-538.

Melzer, K. (2011). Carbohydrate and fat utilization during rest and physical activity. *E-SPEN, the European e-Journal of Clinical Nutrition and Metabolism, 6*(2), e45-e52.

Merrill, E. P., Kramer, F. M., Cardello, A., & Schutz, H. (2002). A comparison of satiety measures. *Appetite, 39*(2), 181-184.

Monsivais, P., & Drewnowski, A. (2007). The rising cost of low-energy-density foods. *Journal of the American Dietetic Association, 107*(12), 2071-2076.

Morio, B., Montaurier, C., Pickering, G., Ritz, P., Fellmann, N., Coudert, J., et al. (1998). Effects of 14 weeks of progressive endurance training on energy expenditure in elderly people. *British Journal of Nutrition, 80*(06), 511-519.

Morland, K., Diez Roux, A. V., & Wing, S. (2006). Supermarkets, other food stores, and obesity: The atherosclerosis risk in communities study. *American Journal of Preventive Medicine, 30*(4), 333-339.

Mrosovsky, N., & Powley, T. L. (1977). Set points for body weight and fat. *Behavioral Biology, 20*(2), 205-223.

Mullineaux, D. R., Barnes, C. A., & Batterham, A. M. (1999). Assessment of bias in comparing measurements: A reliability example. *Measurement in Physical Education and Exercise Science, 3*(4), 195-205.

Murgatroyd, P., Goldberg, G., Leahy, F., Gilsenan, M., & Prentice, A. (1999). Effects of inactivity and diet composition on human energy balance. *International Journal of Obesity, 23*(12), 1269-1275.

Murphy, K. G., & Bloom, S. R. (2006). Review article gut hormones and the regulation of energy homeostasis. *Nature, 444*, 854-859.

Murphy, K. G., & Bloom, S. R. (2004). Gut hormones in the control of appetite. *Experimental Physiology, 89*(5), 507-516.

Mustelin, L., Joutsi, J., Latvala, A., Pietiläinen, K. H., Rissanen, A., & Kaprio, J. (2012). Genetic influences on physical activity in young adults. A twin study. *Medicine & Science in Sports & Exercise, 44*(7), 1293-1301.

Naessen, S., Carlström, K., Byström, B., Pierre, Y., & Lindén Hirschberg, A. (2007). Effects of an antiandrogenic oral contraceptive on appetite and eating behavior in bulimic women. *Psychoneuroendocrinology, 32*(5), 548-554.

Neal, D. T., Wood, W., & Quinn, J. M. (2006). Habits—A repeat performance. *Current Directions in Psychological Science, 15*(4), 198-202.

Neal, D. T., Wood, W., Wu, M., & Kurlander, D. (2011). The pull of the past when do habits persist despite conflict with motives? *Personality and Social Psychology Bulletin, 37*(11), 1428-1437.

Norgan, N., & Durnin, J. (1980). The effect of 6 weeks of overfeeding on the body weight, body composition, and energy metabolism of young men. *The American Journal of Clinical Nutrition, 33*(5), 978-988.

Norton, G., Anderson, A., & Hetherington, M. (2006). Volume and variety: Relative effects on food intake. *Physiology & Behavior, 87*(4), 714-722.

Nybo, L. (2008). Hyperthermia and fatigue. *Journal of Applied Physiology, 104*(3), 871-878.

O'Brien, E., Asmar, R., Beilin, L., Imai, Y., Mancia, G., Mengden, T., et al. (2005). Practice guidelines of the european society of hypertension for clinic, ambulatory and self blood pressure measurement. *Journal of Hypertension, 23*(4), 697.

Office for National Statistics. (2009). Opinions survey report no. 41: Contraception and sexual heath, 2008/09 at *http://www.ons.gov.uk/ons/rel/lifestyles/contraception-and-sexual-health/2008-09/2008-09.pdf*

Ortega, F. B., Artero, E. G., Ruiz, J. R., Vicente-Rodriguez, G., Bergman, P., Hagströmer, M., et al. (2008). Reliability of health-related physical fitness tests in european adolescents. the HELENA study. *International Journal of Obesity, 32*, S49-S57.

Palmiter, R. D. (2007). Is dopamine a physiologically relevant mediator of feeding behavior? *Trends in Neurosciences, 30*(8), 375-381.

Parikh, P., Mochari, H., & Mosca, L. (2009). Clinical utility of a fingerstick technology to identify individuals with abnormal blood lipids and high-sensitivity C-reactive protein levels. *American Journal of Health Promotion, 23*(4), 279-282.

Pecina, S. (2008). Opioid reward 'liking' and 'wanting' in the nucleus accumbens. *Physiology & Behavior, 94*(5), 675-680.

Pérusse, L., Tremblay, A., Leblanc, C., & Bouchard, C. (1989). Genetic and environmental influences on level of habitual physical activity and exercise participation. *American Journal of Epidemiology, 129*(5), 1012-1022.

Perri, M. G., Anton, S. D., Durning, P. E., Ketterson, T. U., Sydeman, S. J., Berlant, N. E., ... & Martin, A. D. (2002). Adherence to exercise prescriptions: effects of prescribing moderate versus higher levels of intensity and frequency. *Health Psychology, 21*(5), 452

Pi-Sunyer, F. X., & Woo, R. (1985). Effect of exercise on food intake in human subjects. *The American Journal of Clinical Nutrition, 42*(5), 983-990.

Pomerleau, M., Imbeault, P., Parker, T., & Doucet, E. (2004). Effects of exercise intensity on food intake and appetite in women. *American Journal of Clinical Nutrition, 80*(5), 1230-1236.

Pories, W. J. (2008). Bariatric surgery: Risks and rewards. *Journal of Clinical Endocrinology & Metabolism, 93*(11 Supplement 1), S89-S96.

Porter, R. (1999). *The greatest benefit to mankind: A medical history of humanity.* WW Norton & Company.

Poston II, W., & Foreyt, J. P. (2000). Successful management of the obese patient. *American Family Physician, 61*(12), 3615-3622.

Powell, K. E., Paluch, A. E., & Blair, S. N. (2011). Physical activity for health: What kind? How much? How intense? On top of what? *Public Health, 32*(1), 349-365.

Prince, S., Adamo, K., Hamel, M., Hardt, J., Gorber, S., & Tremblay, M. (2008). A comparison of direct versus self-report measures for assessing physical activity in adults: A systematic review. *International Journal of Behavioral Nutrition and Physical Activity, 5*(1), 56.

Provencher, V., Drapeau, V., Tremblay, A., Després, J., & Lemieux, S. (2003). Eating behaviors and indexes of body composition in men and women from the Quebec family study. *Obesity Research, 11*(6), 783-792.

Racette, S. B., Schoeller, D. A., Kushner, R. F., Neil, K. M., & Herling-Iaffaldano, K. (1995). Effects of aerobic exercise and dietary carbohydrate on energy expenditure and body composition during weight reduction in obese women. *The American Journal of Clinical Nutrition, 61*(3), 486-494.

Rasmussen, P., Dawson, E. A., Nybo, L., Van Lieshout, J. J., Secher, N. H., & Gjedde, A. (2006). Capillary-oxygenation-level-dependent near-infrared spectrometry in frontal lobe of humans. *Journal of Cerebral Blood Flow & Metabolism, 27*(5), 1082-1093.

Ravussin, E., & Bogardus, C. (1989). Relationship of genetics, age, and physical fitness to daily energy expenditure and fuel utilization. *Am J Clin Nutr, 49*(5), 968-975.

Reger, W. E., Allison, T. G., & Kurucz, R. L. (1984). Exercise, post-exercise metabolic rate and appetite. In F. L. Katch (Ed.), *The 1984 Olympic Scientific Congress Proceedings, vol. 2: Sport, Health, and Nutrition.* (pp. 115-23). Champaign, IL: Human Kinetics Publishers, Inc.

Rennie, K. L., Hennings, S. J., Mitchell, J., & Wareham, N. J. (2001). Estimating energy expenditure by heart-rate monitoring without individual calibration. *Medicine & Science in Sports & Exercise, 33*(6), 939-945.

Rhodes, R., & Smith, N. (2006). Personality correlates of physical activity: A review and meta-analysis. *British Journal of Sports Medicine, 40*(12), 958-965.

Rogers, P. J. (1999). Eating habits and appetite control: A psychobiological perspective. *Proceedings of the Nutrition Society, 58*(01), 59-67.

Rolls, E. T. (2012). Taste, olfactory and food texture reward processing in the brain and the control of appetite. *Proceedings of the Nutrition Society, 71*(4), 488-501.

Rosenkilde, M., Auerbach, P., Reichkendler, M. H., Ploug, T., Stallknecht, B. M., & Sjödin, A. (2012). Body fat loss and compensatory mechanisms in response to different doses of aerobic exercise—a randomized controlled trial in overweight sedentary males. *American Journal of Physiology-Regulatory, Integrative and Comparative Physiology, 303*(6), R571-R579.

Ross, E. Z., Middleton, N., Shave, R., George, K., & Nowicky, A. (2007). Corticomotor excitability contributes to neuromuscular fatigue following marathon running in man. *Experimental Physiology, 92*(2), 417-426.

Rowland, T. W. (1998). The biological basis of physical activity [basic sciences: Brief review]. *Medicine & Science in Sports & Exercise, 30*(3), 392-399.

Sacks, F. M., Bray, G. A., Carey, V. J., Smith, S. R., Ryan, D. H., Anton, S. D., et al. (2009). Comparison of weight-loss diets with different compositions of fat, protein, and carbohydrates. *New England Journal of Medicine, 360*(9), 859-873.

Saelens, B. E., Sallis, J. F., & Frank, L. D. (2003). Environmental correlates of walking and cycling: Findings from the transportation, urban design, and planning literatures. *Annals of Behavioral Medicine, 25*(2), 80-91.

Sakata, I., Tanaka, T., Yamazaki, M., Tanizaki, T., Zheng, Z., & Sakai, T. (2006). Gastric estrogen directly induces ghrelin expression and production in the rat stomach. *Journal of Endocrinology, 190*(3), 749-757.

Saltin, B., & Astrand, P. (1967). Maximal oxygen uptake in athletes. *Journal of Applied Physiology, 23*(3), 353-358.

Santee, J. (2002). Accuracy and precision of the cholestech LDX system in monitoring blood lipid levels. *American Journal of Health-System Pharmacy, 59*, 1774-1779.

Saper, C. B., Chou, T. C., & Elmquist, J. K. (2002). The need to feed: Homeostatic and hedonic control of eating. *Neuron, 36*(2), 199-211.

Schoeller, D. A. (1998). Balancing energy expenditure and body weight. *The American Journal of Clinical Nutrition, 68*(4), 956S-961S.

Schofield, W. (1985). Predicting basal metabolic rate, new standards and review of previous work. *Human Nutrition.Clinical Nutrition, 39*(Suppl. 1), 5-41.

Shaw, K., Gennat, H., O'Rourke, P., & Del Mar, C. (2006). Exercise for overweight or obesity. *Cochrane Database Systematic Reviews,* Issue 4. Art. No.: CD003817. DOI: 10.1002/14651858.CD003817.pub3.

Shepard, T. Y., Weil, K. M., Sharp, T. A., Grunwald, G. K., Bell, M. L., Hill, J. O., et al. (2001). Occasional physical inactivity combined with a high-fat diet may be important in the development and maintenance of obesity in human subjects. *The American Journal of Clinical Nutrition, 73*(4), 703-708.

Shorten, A. L., Wallman, K. E., & Guelfi, K. J. (2009). Acute effect of environmental temperature during exercise on subsequent energy intake in active men. *American Journal of Clinical Nutrition, 90*(5), 1215-1221.

Simonen, R. L., Rankinen, T., Perusse, L., Rice, T., Rao, D., Chagnon, Y., et al. (2003). Genome-wide linkage scan for physical activity levels in the quehec family study. *Medicine & Science in Sports & Exercise, 35*(8), 1355-1359.

Siri, W. E. (1956). The gross composition of the body. *Advances in Biological and Medical Physics, 4*, 239-280.

Sjoedin, A. M., Andersson, A. B., Hoegberg, J. M., & Westerterp, K. R. (1994). Energy balance in cross-country skiers: A study using doubly labeled water. *Medicine & Science in Sports & Exercise, 26*, 720-720.

Small, D. M., Jones-Gotman, M., & Dagher, A. (2003). Feeding-induced dopamine release in dorsal striatum correlates with meal pleasantness ratings in healthy human volunteers. *Neuroimage, 19*(4), 1709-1715.

Smith, J. M., & Ditschun, T. L. (2009). Controlling satiety: How environmental factors influence food intake. *Trends in Food Science & Technology, 20*(6), 271-277.

Snyder, K., Donnelly, J., Jabobsen, D., Hertner, G., & Jakicic, J. (1997). The effects of long-term, moderate intensity, intermittent exercise on aerobic capacity, body composition, blood lipids, insulin and glucose in overweight females. *International Journal of Obesity, 21*(12), 1180-1189.

Solinas, M., Goldberg, S. R., & Piomelli, D. (2008). The endocannabinoid system in brain reward processes. *British Journal of Pharmacology, 154*(2), 369-383.

Spanagel, R., & Weiss, F. (1999). The dopamine hypothesis of reward: Past and current status. *Trends in Neurosciences, 22*(11), 521-527.

Speakman, J. R. (2007). A nonadaptive scenario explaining the genetic predisposition to obesity: The "predation release" hypothesis. *Cell Metabolism, 6*(1), 5-12.

Speth, J. D., & Spielmann, K. A. (1983). Energy source, protein metabolism, and hunter-gatherer subsistence strategies. *Journal of Anthropological Archaeology, 2*(1), 1-31.

Spiegelman, B. M., & Flier, J. S. (2001). Obesity and the regulation review of energy balance. *Cell, 104*, 531-543.

Staten, M. (1991). The effect of exercise on food intake in men and women. *American Journal of Clinical Nutrition, 53*(1), 27-31.

Stensel, D. (2011). Exercise, appetite and appetite-regulating hormones: implications for food intake and weight control. *Annals of Nutrition and Metabolism, 57*(Suppl. 2), 36-42.

Story, M., Kaphingst, K. M., Robinson-O'Brien, R., & Glanz, K. (2008). Creating healthy food and eating environments: Policy and environmental approaches. *Annual Review of Public Health, 29*, 253-272.

St-Pierre, D., Faraj, M., Karelis, A., Conus, F., Henry, J., St-Onge, M., et al. (2006). Lifestyle behaviours and components of energy balance as independent predictors of ghrelin and adiponectin in young non-obese women. *Diabetes & Metabolism, 32*(2), 131-139.

Stroebele, N., & de Castro, J. M. (2006). Influence of physiological and subjective arousal on food intake in humans. *Nutrition, 22*(10), 996-1004.

Stubbe, J. H., Boomsma, D. I., & De Geus, E. (2005). Sports participation during adolescence: A shift from environmental to genetic factors. *Medicine & Science in Sports & Exercise, 37*(4), 563-570.

Stubbe, J. H., Boomsma, D. I., Vink, J. M., Cornes, B. K., Martin, N. G., Skytthe, A., et al. (2006). Genetic influences on exercise participation in 37.051 twin pairs from seven countries. *PLoS One, 1*(1), e22.

Stubbs, R. J., Hughes, D. A., Johnstone, A. M., Horgan, G. W., King, N., & Blundell, J. E. (2004a). A decrease in physical activity affects appetite, energy, and nutrient balance in lean men feeding ad libitum. *American Journal of Clinical Nutrition, 79*(1), 62-69.

Stubbs, R., Hughes, D., Johnstone, A., Whybrow, S., Horgan, G., King, N., et al. (2004b). Rate and extent of compensatory changes in energy intake and expenditure in response to altered exercise and diet composition in humans. *American Journal of Physiology- Regulatory, Integrative and Comparative Physiology, 286*(2), R350-R358.

Stubbs, R., Sepp, A., Hughes, D., Johnstone, A., Horgan, G., King, N., et al. (2002a). The effect of graded levels of exercise on energy intake and balance in free-living men, consuming their normal diet. *European Journal of Clinical Nutrition, 56*(2), 129-140.

Stubbs, R., Sepp, A., Hughes, D., Johnstone, A., King, N., Horgan, G., et al. (2002b). The effect of graded levels of exercise on energy intake and balance in free-living women. *International Journal of Obesity, 26*(6), 866-869.

Sutton, R. (2012). Chapter 10: Adult anthropometric measures, overweight and obesity. In R. Craig, & J. Mindell (Eds.), *Health Survey for England 2011: Volume 1: Health, social care and lifestyles*. Health and Social Care Information Centre, Leeds.

Suzuki, S., Urata, G., Ishida, Y., Kanehisa, H., & Yamamura, M. (1998). Influences of low intensity exercise on body composition, food intake and aerobic power of sedentary young females. *Applied Human Science, 17*(6), 259-266.

Swinburn, B. A., Sacks, G., Hall, K. D., McPherson, K., Finegood, D. T., Moodie, M. L., et al. (2011). The global obesity pandemic: Shaped by global drivers and local environments. *The Lancet, 378*(9793), 804-814.

Swinburn, B., & Ravussin, E. (1993). Energy balance or fat balance? *The American Journal of Clinical Nutrition, 57*(5), 766S-770S.

Teixeira, P., Patrick, H., & Mata, J. (2011). Why we eat what we eat: The role of autonomous motivation in eating behaviour regulation. *Nutrition Bulletin, 36*(1), 102-107.

Thompson, D. A., Wolfe, L. A., & Eikelboom, R. (1988). Acute effects of exercise intensity on appetite in young men. *Medicine & Science in Sports & Exercise, 20*(3), 222-227.

Thompson, D., Batterham, A. M., Bock, S., Robson, C., & Stokes, K. (2006). Assessment of low-to-moderate intensity physical activity thermogenesis in young

adults using synchronized heart rate and accelerometry with branched-equation modeling. *Journal of Nutrition, 136*(4), 1037-1042.

Thompson, J. L. (2009). Exercise in improving health v. performance. *Proceedings of the Nutrition Society, 68*(1), 29-33.

Tipton, C. M. (2008). Susruta of india, an unrecognized contributor to the history of exercise physiology. *Journal of Applied Physiology, 104*(6), 1553-1556.

Tremblay, A. T. A., & Therrien, F. T. F. (2006). Physical activity and body functionality: Implications for obesity prevention and treatment. *Canadian Journal of Physiology and Pharmacology, 84*(2), 149-156.

Tsofliou, F., Pitsiladis, Y., Malkova, D., Wallace, A., & Lean, M. (2003). Moderate physical activity permits acute coupling between serum leptin and appetite–satiety measures in obese women. *International Journal of Obesity, 27*(11), 1332-1339.

Tucci, S., Murphy, L., Boyland, E., Dye, L., & Halford, J. (2010). Oral contraceptive effects on food choice during the follicular and luteal phase of the menstrual cycle: A laboratory based study. *Appetite, 55*(3), 388-392.

Turner, J. E., Markovitch, D., Betts, J. A., & Thompson, D. (2010). Nonprescribed physical activity energy expenditure is maintained with structured exercise and implicates a compensatory increase in energy intake. *American Journal of Clinical Nutrition, 92*(5), 1009-1016.

Ueda, S., Miyamoto, T., Nakahara, H., Shishido, T., Usui, T., Katsura, Y., et al. (2013). Effects of exercise training on gut hormone levels after a single bout of exercise in middle-aged japanese women. *Springerplus, 2*(1), 83-90.

Ueda, S., Yoshikawa, T., Katsura, Y., Usui, T., & Fujimoto, S. (2009a). Comparable effects of moderate intensity exercise on changes in anorectic gut hormone levels and energy intake to high intensity exercise. *Journal of Endocrinology, 203*(3), 357-364.

Ueda, S., Yoshikawa, T., Katsura, Y., Usui, T., Nakao, H., & Fujimoto, S. (2009b). Changes in gut hormone levels and negative energy balance during aerobic exercise in obese young males. *Journal of Endocrinology, 201*(1), 151-159.

Ulijaszek, S. J., & Kerr, D. A. (1999). Anthropometric measurement error and the assessment of nutritional status. *British Journal of Nutrition, 82*(03), 165-177.

Unick, J. L., Otto, A. D., Goodpaster, B. H., Helsel, D. L., Pellegrini, C. A., & Jakicic, J. M. (2010). Acute effect of walking on energy intake in overweight/obese women. *Appetite, 55*, 413-419.

United Nations Department of Economic and Social Affairs. (2009). World contraceptive use at *http://www.un.org/esa/population/publications/contraceptive 2009/contracept2009_wallchart_front.pdf*

Valanou, E., Bamia, C., & Trichopoulou, A. (2006). Methodology of physical-activity and energy-expenditure assessment: A review. *Journal of Public Health, 14*(2), 58-65.

Van Etten, L. M. L. A., Westerterp, K. R., Verstappen, F. T. J., Boon, B. J. B., & Saris, W. H. M. (1997). Effect of an 18-wk weight-training program on energy expenditure and physical activity. *Journal of Applied Physiology, 82*(1), 298-304.

Van Vliet, H., Grimes, D., Lopez, L., Schulz, K., & Helmerhorst, F. (2011). Triphasic versus monophasic oral contraceptives for contraception (review). *Cochrane Database of Systematic Reviews,* Issue 11. Art. No.: CD003553. DOI: 10.1002/14651858. CD003553.pub3.

Van Walleghen, E., Orr, J., Gentile, C., Davy, K., & Davy, B. (2007). Habitual physical activity differentially affects acute and short-term energy intake regulation in young and older adults. *International Journal of Obesity, 31*(8), 1277-1285.

Vatansever-Ozen, S., Tiryaki-Sonmez, G., Bugdayci, G., & Ozen, G. (2011). The effects of exercise on food intake and hunger: Relationship with acylated ghrelin and leptin. *Journal of Sports Science and Medicine, 10*, 283-291.

Verger, P., Louis-Sylvestre, J., & Lanteaume, M. (1994). Free food choice after acute exercise in men. *Appetite, 22*(2), 159-164.

Vlitos, A. L. P., & Davies, G. J. (1996). Bowel function, food intake and the menstrual cycle. *Nutrition Research Reviews, 9*(01), 111-134.

Volkow, N. D., Wang, G., & Baler, R. D. (2011). Reward, dopamine and the control of food intake: Implications for obesity. *Trends in Cognitive Sciences, 15*(1), 37-46.

Wadden, T. A., Butryn, M. L., & Wilson, C. (2007). Lifestyle modification for the management of obesity. *Gastroenterology, 132*(6), 2226-2238.

Wadden, T. A., Vogt, R. A., Andersen, R. E., Bartlett, S. J., Foster, G. D., Kuehnel, R. H., et al. (1997). Exercise in the treatment of obesity: Effects of four interventions on body composition, resting energy expenditure, appetite, and mood. *Journal of Consulting and Clinical Psychology, 65*(2), 269-277.

Wang, X., & Nicklas, B. J. (2011). Acute impact of moderate-intensity and vigorous-intensity exercise bouts on daily physical activity energy expenditure in postmenopausal women. *Journal of Obesity, 2011.*

Wansink, B. (2004). Environmental factors that increase the food intake and consumption volume of unknowing consumers. *Annual Review of Nutrition, 24*, 455-479.

Warren, J. M., Ekelund, U., Besson, H., Mezzani, A., Geladas, N., & Vanhees, L. (2010). Assessment of physical activity–a review of methodologies with reference to epidemiological research: A report of the exercise physiology section of the european

association of cardiovascular prevention and rehabilitation. *European Journal of Cardiovascular Prevention & Rehabilitation, 17*(2), 127-139.

Webber, J. (2003). Energy balance in obesity. *Proceedings of the Nutrition Society, 62*(02), 539-543.

Weir, J. P. (2005). Quantifying test-retest reliability using the intraclass correlation coefficient. *Journal of Strength and Conditioning Research, 19*(1), 231-240.

Wellman, P. J. (2005). Modulation of eating by central catecholamine systems. *Current Drug Targets, 6*(2), 191-199.

Wen, M., Browning, C. R., & Cagney, K. A. (2007). Neighbourhood deprivation, social capital and regular exercise during adulthood: A multilevel study in Chicago. *Urban Studies, 44*(13), 2651-2671.

Westerterp, K. R. (2001). Limits to sustainable human metabolic rate. *Journal of Experimental Biology, 204*(18), 3183-3187.

Westerterp, K. R. (2003). Impacts of vigorous and non-vigorous activity on daily energy expenditure. *Proceedings of the Nutrition Society, 62*(03), 645-650.

Westerterp, K. R. (2008). Physical activity as determinant of daily energy expenditure. *Physiology & Behavior, 93*(4), 1039-1043.

Westerterp, K. R., Meijer, G. A. L., Janssen, E. M. E., Saris, W. H. M., & Hoor, F. T. (1992). Long-term effect of physical activity on energy balance and body composition. *British Journal of Nutrition, 68*(01), 21-30.

Westerterp-Plantenga, M. (1999). Effects of extreme environments on food intake in human subjects. *Proceedings-Nutrition Society of London, 58*(4), 791-798.

Westerterp-Plantenga, M., Kempen, K., & Saris, W. (1998). Determinants of weight maintenance in women after diet-induced weight reduction. *International Journal of Obesity, 22*(1), 1-6.

Westerterp-Plantenga, M., Verwegen, C., Ijedema, M., Wijckmans, N., & Saris, W. (1997). Acute effects of exercise or sauna on appetite in obese and nonobese men. *Physiology and Behavior, 62*(6), 1345-1354.

White, M. A., Whisenhunt, B. L., Williamson, D. A., Greenway, F. L., & Netemeyer, R. G. (2002). Development and validation of the Food - Craving inventory. *Obesity Research, 10*(2), 107-114.

WHO. (2000). *Obesity: Preventing and managing the global epidemic.* World Health Organization technical report series No. 894, Geneva.

WHO. (2004). Human energy requirements: Report of a joint FAO/WHO/UNU expert consultation. Food and Nutrition technical report series No. 1, Rome.

Whybrow, S., Hughes, D. A., Ritz, P., Johnstone, A. M., Horgan, G. W., King, N., et al. (2008). The effect of an incremental increase in exercise on appetite, eating behaviour and energy balance in lean men and women feeding ad libitum. *British Journal of Nutrition, 100*(05), 1109-1115.

Wing, R. R. (1999). Physical activity in the treatment of the adulthood overweight and obesity: Current evidence and research issues. *Medicine & Science in Sports & Exercise, 31*(Suppl. 11), S547- S552.

Wing, R. R., & Phelan, S. (2005). Long-term weight loss maintenance. *The American Journal of Clinical Nutrition, 82*(1), 222S-225S.

Wing, R. R., Lang, W., Wadden, T. A., Safford, M., Knowler, W. C., Bertoni, A. G., et al. (2011). Benefits of modest weight loss in improving cardiovascular risk factors in overweight and obese individuals with type 2 diabetes. *Diabetes Care, 34*(7), 1481-1486.

Wirtshafter, D., & Davis, J. D. (1977). Set points, settling points, and the control of body weight. *Physiology & Behavior, 19*(1), 75-78.

Wisker, E., & Feldheim, W. (1990). Metabolizable energy of diets low or high in dietary fiber from fruits and vegetables when consumed by humans. *The Journal of Nutrition, 120*(11), 1331-1337.

Wisker, E., Maltz, A., & Feldheim, W. (1988). Metabolizable energy of diets low or high in dietary fiber from cereals when eaten by humans. *The Journal of Nutrition, 118*(8), 945-952.

Woo, R., & Pi-Sunyer, F. X. (1985). Effect of increased physical activity on voluntary intake in lean women. *Metabolism, 34*(9), 836-841.

Woo, R., Garrow, J. S., & Pi-Sunyer, F. X. (1982a). Effect of exercise on spontaneous calorie intake in obesity. *The American Journal of Clinical Nutrition, 36*(3), 470-477.

Woo, R., Garrow, J., & Pi-Sunyer, F. (1982b). Voluntary food intake during prolonged exercise in obese women. *The American Journal of Clinical Nutrition, 36*(3), 478-484.

Woods, S. C., & D'Alessio, D. A. (2008). Central control of body weight and appetite. *Journal of Clinical Endocrinology & Metabolism, 93*(11 Supplement 1), S37-S50.

Wren, A., Seal, L., Cohen, M., Brynes, A., Frost, G., Murphy, K., et al. (2001). Ghrelin enhances appetite and increases food intake in humans. *Journal of Clinical Endocrinology & Metabolism, 86*(12), 5992-5992.

Yeomans, M. (2000). Rating changes over the course of meals: What do they tell us about motivation to eat? *Neuroscience and Biobehavioral Reviews, 24*(2), 249-259.

Yeomans, M. R., Gould, N. J., Leitch, M., & Mobini, S. (2009). Effects of energy density and portion size on development of acquired flavour liking and learned satiety. *Appetite, 52*(2), 469-478.

Yeung, R. R. (1996). The acute effects of exercise on mood state. *Journal of Psychosomatic Research, 40*(2), 123-141.

Zhang, Y., Proenca, R., Maffei, M., Barone, M., Leopold, L., & Friedman, J. M. (1994). Positional cloning of the mouse obese gene and its human homologue. *Nature, 372*(6505), 425-432.

Appendix 1

Faculty of Health and Wellbeing Research Ethics Committee
Sport and Exercise Research Ethics Review Group
Report Form

Principal Investigator: Joel Rocha

Title: Effects of an acute bout of moderate-intensity aerobic exercise on subsequent food intake and energy expenditure.

Checklist:

Application form	*Y*
Informed consent form	*Y*
Participant information sheet	*y*
Risk assessment form	*Y*
Pre-screening form	*Y*
Pre-screening form (under 18)	**n/a**
Collaboration evidence/support	**n/a**
CRB Disclosure certificate	**n/a**

Recommendation:

Acceptable: ***f~^***

Not acceptable, see comments:

Acceptable, but see comments: _____

Comments:

Thank you for providing a written response to the reviewers' comments - your application is now acceptable and you may commence your study.

Signature :. **Date:** **> ©**

Professor Edward Winter, Chair
Faculty of Health and Wellbeing Research Ethics Committee

Please remember that an up-to-date project file must be maintained for the duration of the project and afterwards. The project file might be inspected at any time.

Note: *Approval applies until the anticipated date of completion unless there are changes to the procedures, in which case another application should be made.*

Name of Supervisor: David Broom

Faculty of Health and Wellbeing Research Ethics Committee
Sport and Exercise Research Ethics Review Group
Report Form

Principal Investigator: Joel Rocha

Title: Effects of an acute bout of moderate-intensity aerobic exercise on subsequent food intake and energy expenditure during a three day period in active and sedentary females.

Checklist:

Application form	✓
Informed consent form	✓
Participant information sheet	✓
Risk assessment form	✓
Pre-screening form	✓
Pre-screening form (under 18)	n/a
Collaboration evidence/support	n/a
CRB Disclosure certificate	n/a

Recommendation:

Acceptable:	✓
Not acceptable, see comments:	
Acceptable, but see comments:	

Comments:

Thank you for providing a written response to the comments you received.

Your application is now Acceptable and you may commence your study.

Signature : ..... Date: 04.04.11

David Binney
Chair, Sport and Exercise Research Ethics Review Group

Please remember that an up-to-date project file must be maintained for the duration of the project and afterwards. The project file might be inspected at any time.

Note: *Approval applies until the anticipated date of completion unless there are changes to the procedures, in which case another application should be made.*

Name of Supervisor: Edward Winter/David Broom

Faculty of Health and Wellbeing Research Ethics Committee
Sport and Exercise Research Ethics Review Group
Report Form

Principal Investigator: Joel Rocha

Title: The influence of genetic variation in dopamine signalling on the sustainability of lifestyle inventions.

Checklist:

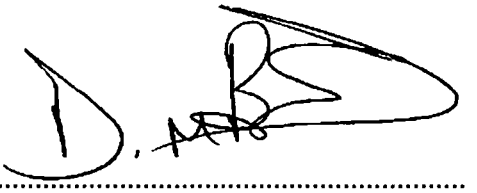
Application form	✓
Informed consent form	✓
Participant information sheet	✓
Risk assessment form	✓
Pre-screening form	✓
Pre-screening form (under 18)	n/a
Collaboration evidence/support	n/a
CRB Disclosure certificate	n/a

Recommendation:

Acceptable:	✓
Not acceptable, see comments:	
Acceptable, but see comments:	

Comments:

Please see attached.



Signature : ..... Date: 06.10.11

David Binney
Chair, Sport and Exercise Research Ethics Review Group

Please remember that an up-to-date project file must be maintained for the duration of the project and afterwards. The project file might be inspected at any time.

Note: *Approval applies until the anticipated date of completion unless there are changes to the procedures, in which case another application should be made.*

Name of Supervisor: David Broom

Appendix 2

L *Sheffield Hallam University*

Faculty of Health and Wellbeing Research Ethics Committee
Sport and Exercise Research Ethics Review Group

Participant Information Sheet

Project Title	Food, Activity and Mood.
Supervisor/Director of Studies	Dr. David Broom, Jenny Paxman, Dr. Caroline Dalton / Professor Edward Winter
Principal Investigator	Joel Rocha
Principal Investigator telephone/mobile number	0114 225 2262

Purpose of Study and Brief Description of Procedures_____

Preliminary Procedures

During the preliminary visit:

- The objectives of the study and its requirements will be explained to you;
- You will be asked to complete a confidential questionnaire regarding your health;
- You will be familiarised with the testing procedures and equipment;
- Any questions you may have will be answered.

Following this you will be asked to complete two exercise tests as follows:

Submaximal cycle erqometer test

You will cycle continuously for 16 min divided into four, 4-min stages. The pedalling rate will be kept constant and at the end of each 4 min stage there will be an increase in the resistance. You will be asked to do the entire test while maintaining a seated position.

Cycle erqometer maximum oxygen uptake (VO^{y}) test

After a 20-30 min recovery period (if you need more time to recover from the submaximal test, an extended period will be allowed) you will begin the cycle ergometer maximum oxygen uptake test. You will have to cycle continuously through 3-min stages until you cannot continue at which point you will give us a signal to stop the test. During the test the pedalling rate will be set constant and at the end of each 3 min stage there will be an increase in the resistance.

Main Trials

There will be two main trials and you will complete both:

- You will complete the cycle ergometer exercise trial and a resting trial at the same time of day and day of week;
- You will start both trials by eating a standard breakfast (within 15 minutes) and resting for one hour. At the end of this resting period a 60 minute exercise or resting period will begin;
- During the cycle ergometer trial you will exercise for 60 minutes at moderate intensity followed by a one hour rest period. After this period we will provide you with a free meal;
- During the resting trial you will be asked to remain seated while working, reading or listening to music during the 3 hours from the end of breakfast until the provided free meal;

Preparation for the tests

Pre-trial recording of your diet

You will be asked to weigh and record everything you eat and drink for **two** days prior to the main trials. You will then consume identical amounts of the same food and drink prior to the next main trial. This is very important in order to control for diet and we will discuss this with you prior to the main trials. No alcohol and caffeine should be consumed on the day before the main trials.

Controlling physical activity

It is crucial to the success of the experiment that you undertake exactly the same type and amount of activity the day before the start of each main trial. Please avoid vigorous activity.

Ten-hour overnight fast

You will finish eating by 11pm on the evenings before the main trials. You may continue to drink water after this time. You will report to the laboratory the following morning without eating breakfast.

Travelling to the laboratory

You should walk in slowly on the morning of each main trial. Please do not run or cycle.

After each main trial

Recording your diet

You will be asked to weigh and record everything you eat and drink for **three** days after both main trials. This time the recordings will not be used to replicate the diet but only for information purposes so you should eat what you want, when you want and as much as you want.

Post-trial recording of physical activity

You will be asked to wear **at all times** a device called Actiheart for the 3 days following the main trials. This is a small, lightweight device (10g) that is supported by just placing 2 small round pads on your chest so it should not affect your daily life in anyway. There is no risk of electrical shock and it is water proof so it should be worn even during the shower.

How much time will it take?

- Preliminary session - **1 hour and 30 minutes;**
- Each main trial: **3 hours.**

You will be encouraged to bring work and reading material with Alternatively you may listen to music.

Possible risks and discomforts

The cycle ergometer maximum oxygen uptake test will cause breat to physical exhaustion but you should recover within a few minutes muscle soreness to a certain degree but is normally of short successfully record heart rate using the Actiheart skin preparation involve cleaning and removing the top layer of the skin on the site going to be placed with an abrasive material. This might induce a sm; and some local redness, however, this is normal and should not Despite rare, the prolonged use of the pads (3 days) may also caus* reaction due to different skin types reacting differently to differ* disappear soon after the removal of the pad.

Benefits of the study

The study will provide important information regarding the effects i activity on mood. We will provide you with feedback on your own resul discuss these with you.

Remember that you always have the right to withdraw from the study a

It has been made clear to me that, should I feel that these Regulations ar that my interests are otherwise being ignored, neglected or denied, I shoi Edward Winter, Chair of the Faculty of Health and Wellbeing Research E 0114 225 4333) who will undertake to investigate rny complaint.

Sheffield Hallam University

Faculty of Health and Wellbeing Research Ethics Committee
Sport and Exercise Research Ethics Review Group

Participant Information Sheet

Project Title	Food, Activity and Mood.
Supervisor/Director of Studies	Dr. David Broom, Jenny Paxman, Dr. Caroline Dalton / Professor Edward Winter
Principal Investigator	Joel Rocha
Principal Investigator telephone/mobile number	0114 225 2262

Purpose of Study and Brief Description of Procedures_____

Preliminary Procedures

During the preliminary visit:

- The objectives of the study and its requirements will be explained to you;
- You will be asked to complete a confidential questionnaire regarding your health;
- You will be familiarised with the testing procedures, equipment and recording techniques;
- Any questions you may have will be answered.

Following this you will be asked to complete two exercise tests as follows:

Submaximal cycling test

You will cycle continuously for 16 min divided into four, 4-min stages. The pedalling rate will be kept constant and at the end of each 4 min stage there will be an increase in the resistance. You will be asked to do the entire test while maintaining a seated position.

Maximal cycling test

After a 20-30 min recovery period (if you need more time to recover from the submaximal test, an extended period will be allowed) you will begin the maximal cycling test. You will have to cycle continuously through 3-min stages until you cannot continue at which point you will give us a signal to stop the test. During the test the pedalling rate will be set constant and at the end of each 3 min stage there will be an increase in the resistance.

Main Trials

There will be two main trials and you will complete both:

- You will complete the exercise trial and a resting trial at the same time of day and day of week separated by approximately a 4 week interval;
- You will start both trials by eating a standard breakfast (within 15 minutes) and resting for one hour. At the end of this resting period a 60 minute exercise or resting period will begin;
- During the cycling trial you will exercise for 60 minutes at moderate intensity followed by a one hour rest period. After this period we will provide you with a free meal;
- During the resting trial you will be asked to remain seated while working, reading or listening to music during the 3 hours from the end of breakfast until the provided free meal;

Preparation for the tests

Pre-trial recording of your diet

You will be asked to weigh and record everything you eat and drink for **two** days prior to the main trials. You will then consume identical amounts of the same food and drink prior to the next main trial. This is very important in order to control for diet and we will discuss this with you prior to the main trials. No alcohol and caffeine should be consumed on the day before the main trials.

Controlling physical activity

It is crucial to the success of the experiment that you undertake exactly the same type and amount of activity the day before the start of each main trial. Please avoid vigorous activity.

Ten-hour overnight fast

You will finish eating by 11pm on the evenings before the main trials. You may continue to drink water after this time. You will report to the laboratory the following morning without eating breakfast.

Travelling to the laboratory

You should walk in slowly on the morning of each main trial. Please do not run or cycle.

After each main trial

Recording your diet

You will be asked to weigh and record everything you eat and drink for **three** days after both main trials. This time the recordings will not be used to replicate the diet but only for information purposes so you should eat what you want, when you want and as much as you want.

Post-trial recording of physical activity

You will be asked to wear **at all times** a device called Actiheart for the 3 days following the main trials. This is a small, lightweight device (10g) that is supported by just placing 2 small circular pads on your chest so it should not affect your daily life in anyway. There is no risk of electrical shock and it is water proof so it should be worn even during the shower.

How much time will it take?

- Preliminary session - **1 hour 30 minutes;**
- Each main trial: **3 hours.**

You will be encouraged to bring work and reading material with you for the main trials. Alternatively you may listen to music.

Possible risks and discomforts

The maximal cycling test will cause breathlessness and will lead to physical exhaustion but you should recover within a few minutes. You might experience muscle soreness to a certain degree but is normally of short duration. In order to successfully record heart rate using the Actiheart skin preparation is necessary. This will involve cleaning and removing the top layer of the skin on the sites where the pads are going to be placed with an abrasive material. This might induce a small degree of discomfort and some local redness, however, this is normal and should not remain for very long. Despite rare, the prolonged use of the pads (3 days) may also cause in some cases a skin reaction due to different skin types reacting differently to different pads but this will disappear soon after the removal of the pad.

Benefits of the study

The study will provide important information regarding the effects of food and moderate activity on mood. We will provide you with feedback on your own results and will be happy to discuss these with you.

Remember that you always have the right to withdraw from the study at any time.

It has been made clear to me that, should I feel that these Regulations are being infringed or that my interests are otherwise being ignored, neglected or denied, I should inform Mr David Binney, Chair of the Faculty of Health and Wellbeing Research Ethics Committee (Tel: 0114 225 5679) who will undertake tojnvestigate my complaint_____

! *Sheffield Hallam University*

Faculty of Health and Wellbeing Research Ethics Committee
Sport and Exercise Research Ethics Review Group

Participant Information Sheet

Project Title	Exercise, Food, Genetics and Mood.
Supervisor/Director of Studies	Dr. David Broom and Dr. Caroline Dalton / Professor Edward Winter
Principal Investigator	Joel Rocha
Principal Investigator telephone/mobile number	0114 225 2262

Purpose of Study and Brief Description of Procedures_____

You are being invited to take part in a research study. Before you decide if you wish to take part it is important for you to understand why the research is being undertaken and what it will involve for you. Please take time to read the following information carefully. One of our team will go through this information sheet with you and answer any questions you have. Please ask if there is anything that is not clear or if you would like more information. Take your time to decide if you would like to take part.

Each participant will attend two preliminary sessions consisting of one exercise test and health screening;

Preliminary Procedures of First Preliminary Trial

- Explanation of the study objectives and its requirements;
- Completion of a pre-test medical questionnaire;
- Completion of Eating and Activity Questionnaires;
- Tour of the University facilities to be used during the study;
- Answer all participants' questions and doubts.

Preliminary Procedures of Second Preliminary Trial

- Habituate participants with the testing procedures and equipment;
- Habituate participants with dietary recording and the use of the actiheart;
- Completion of the Astrand Bike Test and anthropometric measurements;
- Completion of a cheek DNA sample. The process to take the cheek cell sample is entirely painless; it is very similar to gently brushing the inside of your cheek with a toothbrush. You would carry out this process yourself with help from our researcher;
- Answer all participants' questions and doubts.

Submaximal friction-braked cycle ergometer test (The Astrand Bike Test)

All participants will undertake the Astrand Bike Test (Astrand & Ryhming, 1954) which is a 6 minute cycling test that can be used in men of various ages who are not used to exercising as it involves cycling from low to moderate intensity.

Baseline Week

During your baseline week you will:

- Record all of the food that you eat for 1 week;
- Wear a device that will record the amount of activity that you undertake;
- Give a blood sample - Staff taking the sample are fully trained and whilst there will be some initial pain felt as a small scratch on the skin taking blood is a routine procedure.

Intervention

You will then undertake a 12 week exercise intervention.

12 week exercise intervention

Each exercise session will involve a 1 hour activity block consisting of 5-10 minute warm up, 40 minute main exercise and 5-10 minute cool down. Numerous exercise modes will be available including treadmill running, cycling, rowing and stepping and intensity will be « 60% of heart rate reserve which is an intensity that will make you become warmer and breath harder than normal. At least 3 days per week sessions will be supervised in the exercise suite based at Collegiate Hall, Collegiate Crescent Campus Sheffield Hallam University where you will be required to wear a heart rate monitor. You will also be offered the opportunity to undertake a further 2 unsupervised sessions but the opportunity will be provided for these to be supervised if necessary. As the study progresses more sessions will become unsupervised and activity undertaken recorded.

Post Intervention Week

Everything that was undertaken in the baseline week (see above) will be measured again in the post intervention week.

Possible risks and discomforts

In order to successfully record heart rate using the Actiheart skin preparation is necessary. This will involve cleaning and removing the top layer of the skin on the sites where the pads are going to be placed with an abrasive material. This might induce a small degree of discomfort and some local redness, however, this is normal and should not remain for very long. Despite rare, the prolonged use of the pads (7 days) may also cause in some cases a skin reaction due to different skin types reacting differently to different pads but this will disappear soon after the removal of the pad. As previously mentioned you will feel an initial scratch on the skin when the blood sample is being taken but the pain will soon subside.

Benefits of the study

The study will provide important information regarding the effects of exercise and food on mood. We will provide you with feedback on your own results and will be happy to discuss these with you.

What will happen to the results at the end of the study?_____

The results will be published as academic reports in scientific journals. There will be no identifying information published. We will also write a summary report of anything we discover which we will send to any of the people who participated in the study who want to see it. This will not contain any identifying information.

Will my taking part in this study be kept confidential?

Yes, no one will know you have taken part in the study except the members of the research team. We will not inform your G.P. unless you ask us to.

Has this study got ethical approval?

Yes, this study was approved by the local Ethics Committee. This means that experts who are not involved in the study have carefully considered the aims and methods of the study and agreed that they meet their guidelines.

Right to Withdraw

Remember that you always have the right to withdraw from the study at any time.

It has been made clear to me that, should I feel that these Regulations are being infringed or that my interests are otherwise being ignored, neglected or denied, I should inform Mr David Binney, Chair of the Faculty of Health and Wellbeing Research Ethics Committee (Tel: 0114 225 5679) who will undertake to investigate my complaint. _____ __ _____

Appendix 3

I Sheffield Hallam University

Faculty of Health and Wellbeing Research Ethics Committee
Sport and Exercise Research Ethics Review Group

INFORMED CONSENT FORM

TITLE OF PROJECT: Food, Activity and Mood.

The participant should complete the whole of this sheet himself

Have you read the Participant Information Sheet?	YES/NO
Have you had an opportunity to ask questions and discuss this study?	YES/NO
Have you received satisfactory answers to all of your questions?	YES/NO
Have you received enough information about the study?	YES/NO

To whom have you spoken?

Do you understand that you are free to withdraw from the study:

- at any time
- without having to give a reason for withdrawing
- and without affecting your future medical care YES/NO

Have you had sufficient time to consider the nature of this project? YES/NO

Do you agree to take part in this study? YES/NO

Signed..... Date.....

(NAME IN BLOCK LETTERS).....

Appendix 4

L *Sheffield Hallam University*

Faculty of Health and Wellbeing Research Ethics Committee
Sport and Exercise Research Ethics Review Group

Pre-Test Medical Questionnaire

Name:_____

Date of Birth:_____ Age:_____Sex:_____

Telephone/Mobile:_____E-mail:_____

Please answer the following questions by putting a circle round the appropriate response or filling in the blank.

1. How would you describe your present level of activity?
   Sedentary / Moderately active / Active / Highly active

2. How would you describeyou present level of fitness?
   Unfit / Moderately fit / Trained / Highly trained

3. How would you consider your present body weight?
   Underweight / Ideal / Slightly over / Very overweight

4. Didyour weight fluctuate in the past 6 months? Yes / No

   4.1 If yes, how much?.....

5. Are you currently trying to lose or gain weight? Yes / No

   5.1 If yes, how?.....

6. Smoking Habits

Are you currently a smoker?	Yes / No
How many do you smoke	..... per day
Are you a previous smoker?	Yes / No
How long isit since youstopped?	..... years
Were you an occasional smoker?	Yes / No per day
Were you a regular smoker?	Yes / No per day

7. Do you drink alcohol? Yes / No
   If you answered **Yes,** do you usually have?
   An occasional drink / a drink every day / more than one drink a day?

8. Have you had to consult your doctor within the last six months? Yes / No

If you answered **Yes,** please give details.....

9. Are you currently menstruating? .....

9.1 Dates of last menstruation.....

9.2 Do you have regular menstrual cycles (between 21-35 days)?

9.3 Are you taking birth control pills / oestrogen pills?.....
If you answered **Yes,** please give details.....

9.4 Do you use any other contraceptive hormonal preparation? ...
If you answered **Yes,** please give details.....

9.5 Do you usually experience any premenstrual symptoms?
If you answered **Yes,** what are your symptoms?.....

10. Are you presently taking any form of medication? Yes / No
If you answered **Yes,** please give details.....

11. As far as you are aware, do you suffer or have you ever suffered from:

a Diabetes?	Yes / No	**b** Asthma?	Yes / No
c Epilepsy?	Yes / No	**d** Bronchitis?	Yes / No
e *Any form of heartcomplaint?	Yes / No	**f** Raynaud's Disease?	Yes / No
g *Marfan's Syndrome?	Yes / No	**h** *Aneurysm/embolism?	Yes / No
I Anaemia	Yes / No		

12. *Is there a history of heart disease in your family? Yes/No

13. *Do you currently have any form of muscle or joint injury? Yes / No
If you answered **Yes,** please give details.....

14. Have you had to suspend your normal training in the last two weeks? Yes / No
If the answer is **Yes** please give details.....

If blood is not being taken from you please disregard Section 14. below.

15. * Please read the following questions:

a)	Are you suffering from any known serious infection?	Yes / No
b)	Have you had jaundice within the previous year?	Yes / No
c)	Have you ever had any form of hepatitis?	Yes / No
d)	Are you HIV antibody positive	Yes / No
e)	Have you had unprotected sexual intercourse with any person from an HIV high-risk population?	Yes / No
f)	Have you ever been involved in intravenous drug use?	Yes / No
g)	Are you hemophiliac?	Yes / No

16. As far as you are aware, is there anything that might prevent you from successfully completing the tests that have been outlined to you? Yes / No

IF THE ANSWER TO ANY OF THE ABOVE IS YES THEN:

a) Discuss the nature of the problem with the Principal Investigator.

b) Questions indicated by (*) Allow your Doctor to fill out the 'Doctors Consent Form provided.

As far as I am aware the information I have given is accurate.

Signature: .....

Date:..../...../.....

Appendix 5

PHYSICAL ACTIVITY QUESTIONNAIRE

During one week, how many times on average do you do the following kinds of activities?

(a) **High intensity** (heart beats rapidly)
For example: running, squash, hockey, football, volleyball, vigorous swimming, vigorous long distance cycling.

times per week. Average time per session

(b) **Moderate intensity** (challenging but not exhausting)
For example: fast walking, light jogging, easy cycling, badminton, easy swimming, dancing.

times per week. Average time per session_____

(c) **Low intensity** (minimal effort)
For example: easy walking, yoga, archery, fishing, bowling, golf.

times per week. Average time per session_____

From all the physical activity you do, how much time is spent undertaking moderate aerobic exercise?

Less than 2.5 hours **Q** 2.5 hours or more j **j**

Signature:

Date:

Appendix 6

TFEQ

Participant ID..... Date

Please answer the following questions by circling the response.

1. I deliberately take small helpings as a means of controlling my weight.

definitely true *mostly true* *mostly false* *definitely false*

2. I consciously hold back at meals in order not to gain weight.

definitely true *mostly true* *mostly false* *definitely false*

3. I do not eat some foods because they make me fat.

definitely true *mostly true* *mostly false* *definitely false*

4. How frequently do you avoid 'stocking up' on tempting foods?

almost never *seldom* *usually* *almost always*

5. How likely are you to consciously eat less than you want?

unlikely *slightly likely* *moderately likely* *very likely*

6. On a scale of 1 to 8, where 1 means no restraint in eating (eating whatever you want, whenever you want it) and 8 means total restraint (constantly limiting food intake and never 'giving in'), what number would you give yourself?

Eat whatever I want, whenever I want it

1
2
3
4
5
6
7

8

Constantly limiting food intake, never 'giving in'

7. When I smell a sizzling steak or a juicy piece of meat, I find it very difficult to keep from eating, even if I have just finished a meal.

definitely true *mostly true* *mostly false* *definitely false*

8. Sometimes when I start eating, I just can't seem to stop.

definitely true *mostly true* *mostly false* *definitely false*

9. Being with someone who is eating often makes me hungry enough to eat also.

definitely true *mostly true* *mostly false* *definitely false*

10. When I see a real delicacy, I often get so hungry that I have to eat right away.

definitely true *mostly true* *mostly false* *definitely false*

11. I get so hungry that my stomach often seems like a bottomless pit.

definitely true *mostly true* *mostly false* *definitely false*

12. I am always hungry so it is hard for me to stop eating before I finish the food on my plate.

definitely true *mostly true* *mostly false* *definitely false*

13. I am always hungry enough to eat at any time.

definitely true *mostly true* *mostly false* *definitely false*

14. How often do you feel hungry?

only at mealtimes *sometimes between meals*

often between meals *almost always*

15. Do you go on eating binges though you are not hungry?

never *rarely* *sometimes* *at least once a week*

16. When I feel anxious, I find myself eating.

definitely true *mostly true* *mostly false* *definitely false*

17. When I feel blue, I often overeat.

definitely true *mostly true* *mostly false* *definitely false*

18. When I feel lonely, I console myself by eating.

definitely true *mostly true* *mostly false* *definitely false*

Appendix 7

Shortened Premenstrual Assessment Form

Participant ID:_____ Date:_____

For each of the symptoms below, circle the number that most closely describes the intensity of your premenstrual symptoms during vour last cycle. These are symptoms that would occur during the premenstrual phase of your cycle. This phase begins about seven days prior to menstrual bleeding (or seven days before your period) and ends about the time bleeding starts. Rate each item on this list on a scale from 1 (not present or no change from usual) to 6 (extreme change, perhaps noticeable even to casual acquaintances).

	1"No change					Extreme changed
1. Pain, tenderness, enlargement or swelling of breasts	1	2	3	4	5	6
2. Feeling unable to cope or overwhelmed by ordinary demands	1	2	3	4	5	6
3. Feeling under stress	1	2	3	4	5	6
4. Outburst of irritability or bad temper	1	2	3	4	5	6
5. Feeling sad or blue	1	2	3	4	5	6
6. Backaches, joint and muscle pain, or joint stiffness	1	2	3	4	5	6
7. Weight gain	1	2	3	4	5	6
8. Relatively steady abdominal heaviness, discomfort or pain	1	2	3	4	5	6
9. Edema, swelling, puPfiness, or water retention	1	2	3	4	5	6
10. Feeling bloated	1	2	3	4	5	6

Total Score

Allen S, et al. The Shortened Premenstrual Assessment form. *J Reprod Med.* **1991;36(l l):769-72.**

Appendix 8

t *Sheffield Hallam University*

Faculty of Health and Wellbeing

Centre for Sport and Exercise Science

Food Declaration

1. During this study you will be given a standard breakfast that will consist in cornflake cereals, semi-skimmed milk and a glass of orange juice. Do you dislike or feel uncomfortable with any of these foods?

YES □ NO □

2. Do you avoid or dislike any of the following ingredients (White Penne Pasta, Vine-ripened Tomatoes, Mascarpone Cheese, Butter, Sunflower Seed Oil, Onions, Sea Salt, Parsley, Garlic, Basil and Black Pepper)?

YES □ NO □

3. Are you currently engaged in any specific diets (e.g. Atkinson's diet) ?

YES □ NO **H**

Signature:

Date: ...../...../

Appendix 9

Participant ID: **Trial:** **Date:**

Visual Analogue Scale

Time.....

Place a vertical mark on the horizontal line corresponding to the intensity of how you feel right now based on the following questions.

How happy do you feel?

Not at all happy Very happy

How tired do you feel?

Not at all tired Very tired

How hungry do you feel?

Not at all hungry Very Hungry

How nervous do you feel?

Not at all nervous Very nervous

How nauseous do you feel?

Not at all nauseous Very nauseous

Appendix 10

Sheffield Hallam University

Faculty of Health and Wellbeing

Centre for Sport and Exercise Science

How to complete this food diary:

1 Be honest and list all of the food and drinks you have consumed throughout the day.

2 Carry this diary with you at all times on these days and complete after every meal, drink or snack.

3 When weighing is not possible give as much detail as possible to show quantity. For example: Teaspoons, tablespoons, cups or pints. Can size. Small, medium or large portion.

4 Be as detailed as possible. For example: Brand names or ingredients of composite meals (or cut out the ingredients list). Type of milk: skimmed, semi-skimmed or full fat. Fried, steamed, boiled, roasted, fresh, frozen, canned or dried food.

5 Use the comments box to explain any unusual symptoms or events in the day.

Joel Rocha

****

Email: J.Rocha@shu.ac.uk

Example of Food Diary Sheet (1 sheet was given per day)

Participant ID: _____ Date: _____

Time awake (hrs):_____ Time asleep (hrs):_____

Time (e.g. 9 am)	**Amount** (e.g. give as tsp, tbsp, cups, oz, weight or portion size)	**Description of food and/or drink** (give a detailed description including type of food brand name, or restaurant)	**Leftovers** (give detailed description of food that was uneaten)	**Mood** (e.g. bored, sad, happy, tired)

Appendix 11

Appendix 12

FOOD CRAVING INVENTORY

Name:_____ Date:_____

Food craving is defined as an intense desire to consume a particular food (or food type) that is difficult to resist.

For each of the foods listed below (Items 1-28), please circle the appropriate letter using the following scale.

Over the past month, how often have you experienced a craving for the food?

A = Never

B = Rarely (once or twice)

C = Sometimes

D = Often

E = Always/almost every day

List of foods	Never	Rarely	Sometimes		Often	Almost everyday
Cakes	A	B	C	D		
Pizza	A	B	C	D		
Fried Chicken	A	B	C	D		
Sausages	A	B	C	D		
French Fries/Chips	A	B	C	D		
Rice	A	B	C	D		
Sausage Rolls	A	B	C	D		
Gravy	A	B	C	D		
Hamburger /Beefburger	A	B	C	D		
Biscuits	A	B	C	D		

List of foods	Never	Rarely	Sometimes	Often	Almost everyday
Ice Cream	A	B	C	D	E
Pasta	A	B	C	D	E
Fried Fish	A	B	C	D	E
Cookies	A	B	C	D	E
Chocolate	A	B	C	D	E
Pancakes/Waffles	A	B	C	D	E
Bread Rolls/ Bagels/Baps	A	B	C	D	E
Doughnuts	A	B	C	D	E
Sweets	A	B	C	D	E
Brownies/Muffins	A	B	C	D	E

List of foods	Never	Rarely	Sometimes	Often	Almost everyday
Bacon	A	B	C	D	E
Steak	A	B	C	D	E
Danish pastry	A	B	C	D	E
Baked Potatoes	A	B	C	D	E
Sponge Cake	A	B	C	D	E
Cereals	A	B	C	D	E
Sandwich Bread	A	B	C	D	E
Crisps	A	B	C	D	E